
**Watershed Evaluation of Beneficial
Management Practices (WEBs) on
Escherichia coli in the
Steppler Watershed, Manitoba, Canada**

By D. J. Green

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Water Science & Management Branch



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SUMMARY

A study was conducted in the Steppler Watershed, Manitoba, Canada from 2004 to 2007 under an Agriculture and Agri-food Canada WEBs (Watershed Evaluation of Beneficial Management Practices) program. The WEBs program was established to evaluate the effectiveness of several Beneficial Management Practices (BMPs) for nutrients, sediment, and bacteria on field and a small watershed scale. The targeted BMP for evaluating mitigating effects on *Escherichia coli* (*E. coli*) was use of a holding pond to intercept and temporarily hold runoff originating from a farmyard-cattle-corral area to prevent *E. coli* and associated organisms in runoff from being transported to downstream receiving surface waters. The WEBs study was conducted with the collaboration of the land owner within the Steppler Watershed and various agencies that included Prairie Farm Rehabilitation Administration, the Deerwood Soil and Water Association, Environment Canada, Manitoba Agriculture, Food and Rural Initiatives, and Manitoba Water Stewardship.

Mean *E. coli* counts from most sites within the Steppler Watershed were usually less than the water quality guideline limit for primary recreational activities in Manitoba that is a maximum of 200 *E. coli* 100 mL⁻¹. Mean *E. coli* counts in runoff from the farmyard-cattle-corral from 2004 to 2007 were significantly greater than this guideline and were usually significantly greater than at other water quality sampling sites in the watershed. Annual maximum counts also came from the farmyard-cattle-corral and were as high as 2,300,000 *E. coli* 100 mL⁻¹ during 2007. The lowest mean *E. coli* counts were generally 10 *E. coli* 100 mL⁻¹ or less and came from a small, natural wooded watershed that was also not sampled as extensively as the Steppler Watershed.

The holding pond was generally effective at intercepting and holding farmyard-cattle-corral runoff highly contaminated with *E. coli* from being transported to further downstream areas, but its effectiveness to significantly reduce counts any more than natural attenuation prior to use of the holding pond was inconclusive. There was no significant difference in mean *E. coli* counts between sites upstream and downstream of runoff from the farmyard-cattle-corral from 2004 to 2007 despite operation of the holding pond in 2006 and 2007. Between-year mean *E. coli* counts for each individual site were also not significantly different. It appeared distances between sampling sites of 500 m or more in the Steppler Watershed provided opportunity for substantial count reductions through additional dilution and/or increased die-off due to exposure to the sunlight's ultraviolet radiation. Still, *E. coli* counts entering the holding pond were often in the range of 10⁴ to 20⁵ *E. coli* 100 mL⁻¹ while counts were one or more orders of magnitude lower when the pond was being emptied. Counts generally declined to less than 200 *E. coli* 100 mL⁻¹ before the pond was empty. It took approximately 14 to 24 days from the time spring runoff effectively stopped filling the holding pond to when *E. coli* counts in the pond were 200 *E. coli* 100 mL⁻¹ or less during 2006 and 2007,

respectively. It took approximately 27 to 37 days from the time spring runoff started filling the holding pond to when *E. coli* counts in the pond were 200 *E. coli* 100 mL⁻¹ or less during 2006 and 2007, respectively.

This study showed that frequency and magnitude of runoff events could be major factors on effectiveness to operate the holding pond. Significant increases in *E. coli* counts entering the holding pond from the farmyard-cattle-corral generally coincided with runoff of 2 L s⁻¹ or more. High runoff volumes over a short period of time could quickly fill the pond and cause some overflow of effluent that had a minimal amount of treatment out an emergency spillway. Periodic runoff events following the initial main spring runoff period could also provide fresh *E. coli* contributions that affected how quickly within pond counts were reduced. However, it appeared that increases in holding pond counts from these runoff events were more minor if the volume in the holding pond was greater than the runoff volume.

Occasional very high *E. coli* counts occurred in the Stepler Watershed at some sampling sites other than from the farmyard-cattle-corral. These incidences appeared to often coincide with the presence of livestock nearby these sites during runoff events. Presence of cattle on occasion nearby sampling sites upstream and downstream of the holding pond runoff was a confounding factor that obscured determining the effectiveness of the holding pond on *E. coli* count reduction to downstream waters. The observations during this study indicated how important timing and location for livestock management would be to maintain effectiveness of some BMPs.

There was no apparent effect or reduction to *E. coli* counts at irrigation ejector nozzles when the holding pond was being emptied via a small irrigation pumping system with an estimated pumping pressure of 90 pounds per square inch.

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RÉSUMÉ

De 2004 à 2007, Agriculture et Agroalimentaire Canada a réalisé une étude dans le bassin hydrographique de Steppeler de la province du Manitoba au Canada dans le cadre de son programme d'évaluation des pratiques de gestion bénéfiques à l'échelle des bassins hydrographiques. Ce programme a été conçu pour évaluer l'efficacité de plusieurs pratiques de gestion bénéfiques en ce qui concerne les nutriments, les sédiments et les bactéries, à l'échelle des champs et des petits bassins hydrographiques. L'étude portait sur l'évaluation des effets de réduction des concentrations d'*Escherichia coli* (*E. coli*) d'une pratique de gestion bénéfique particulière, soit l'utilisation d'un étang de retenue pour intercepter et temporairement retenir le ruissellement provenant d'une aire agricole et d'élevage de bétail afin d'empêcher que les bactéries *E. coli* et autres organismes connexes soient transportés dans les eaux de surface en aval. L'étude a été réalisée en collaboration avec le propriétaire des terres situées dans le bassin hydrographique Steppeler et avec différents organismes, notamment Administration du rétablissement agricole des Prairies, la Deerwood Soil and Water Management Association, Environnement Canada, Agriculture, Alimentation et Initiatives rurales Manitoba et Gestion des ressources hydriques Manitoba.

Les concentrations moyennes d'*E. coli* dans la plupart des sites situés dans le bassin hydrographique Steppeler étaient habituellement moins élevées que la limite recommandée au Manitoba concernant la qualité des eaux utilisées principalement à des fins récréatives, qui est d'un maximum de 200 par 100 ml d'eau. De 2004 à 2007, les concentrations moyennes d'*E. coli* dans le ruissellement provenant de l'aire agricole et d'élevage de bétail étaient considérablement plus élevées que la limite recommandée et étaient habituellement beaucoup plus élevées que celles des échantillons prélevés dans d'autres sites situés dans le bassin. Les concentrations annuelles maximales provenaient aussi de l'aire agricole et d'élevage de bétail et ont atteint jusqu'à 2 300 000 *E. coli* par 100 ml d'eau en 2007. Les concentrations moyennes les moins élevées étaient habituellement de 10 *E. coli* ou moins par 100 ml d'eau, et provenaient d'un petit bassin hydrologique naturel et boisé qui n'a pas fait l'objet d'un échantillonnage aussi considérable que celui de Steppeler.

En général, l'étang de retenue était efficace pour intercepter le ruissellement provenant de l'aire agricole et d'élevage de bétail, hautement contaminé par l'*E. coli*, et l'empêcher de se répandre dans des zones situées plus loin en aval. Toutefois, son efficacité à réduire les concentrations de façon beaucoup plus importante que l'atténuation naturelle qui avait lieu avant la mise en place de l'étang de retenue n'est pas concluante. On n'a pas enregistré de différences importantes dans les concentrations moyennes des sites en aval et en amont du ruissellement provenant de l'aire agricole et d'élevage de bétail de 2004 à 2007 malgré l'utilisation de l'étang de retenue en 2006 et en 2007. En outre, la différence entre années des concentrations moyennes d'*E. coli* pour

chaque site individuel n'était pas significative. Il s'est avéré qu'une distance de 500 m ou plus entre les sites d'échantillonnage dans le bassin hydrographique permettait une importante réduction des concentrations grâce à une dilution additionnelle ou à une disparition plus accélérée en raison de l'exposition aux rayons ultraviolets du soleil. Malgré cela, les concentrations d'*E. coli* entrant dans l'étang de retenue allaient souvent de 10^4 à 20^5 par 100 ml, alors que pendant le vidage de l'étang, les concentrations étaient au moins un ordre de grandeur moins élevé. Les concentrations diminuaient généralement à moins de 200 *E. coli* par 100 ml d'eau avant que l'étang ne soit vidé. En 2006 et 2007, il a fallu environ 14 et 24 jours respectivement pour que les concentrations d'*E. coli* dans l'étang de retenue diminuent à 200 ou moins par 100 ml d'eau, à partir du moment où le ruissellement du printemps a cessé de remplir l'étang. En 2006 et 2007, il a fallu environ 27 et 37 jours respectivement pour que les concentrations d'*E. coli* dans l'étang de retenue diminuent à 200 ou moins par 100 ml d'eau, à partir du moment où le ruissellement du printemps a commencé à remplir l'étang.

Selon l'étude, la fréquence et l'ampleur des ruissellements peuvent être des facteurs jouant un rôle majeur sur l'efficacité d'un étang de retenue. Les augmentations importantes des concentrations d'*E. coli* provenant de l'aire agricole et d'élevage de bétail et entrant dans l'étang de retenue coïncidaient généralement avec des ruissellements de deux litres ou plus par seconde. Des volumes de ruissellement élevés s'écoulant durant une courte période de temps pouvaient remplir l'étang rapidement et causer des débordements d'effluents qui s'écoulaient par un déversoir d'urgence et qui n'avaient subi qu'un traitement minimal. Les ruissellements périodiques qui suivaient la première période principale de ruissellements du printemps pouvaient aussi apporter des quantités d'*E. coli* non traitées qui touchaient la rapidité avec laquelle les concentrations diminuaient dans l'étang. Toutefois, il s'est avéré que les augmentations de concentrations dans l'étang de retenue qui résultaient de ces ruissellements périodiques étaient moins importantes si le volume contenu dans l'étang était plus élevé que celui des ruissellements.

À l'occasion, on a relevé des concentrations très élevées d'*E. coli* dans le bassin de Steppeler dans des sites d'échantillonnage autres que ceux de l'aire agricole et d'élevage de bétail. Ces circonstances semblaient souvent coïncider avec la présence de bétail à proximité des sites en question lorsque qu'il y avait des ruissellements. La présence occasionnelle de bétail près des sites d'échantillonnage en amont et en aval des ruissellements qui s'écoulaient dans l'étang de retenue constituait une variable de confusion qui a nui à la détermination de l'efficacité de l'étang de retenue sur la réduction des concentrations d'*E. coli* dans les eaux en aval. Les observations menées durant cette étude ont indiqué à quel point la synchronisation et l'emplacement dans la gestion du bétail joueraient un rôle important pour le maintien de l'efficacité de certaines pratiques de gestion bénéfiques.

Il n'y avait aucune répercussion manifeste sur les concentrations d'*E. coli* ni de diminution visible de celles-ci dans les tuyères d'éjecteur lorsque l'étang de retenue était vidé au moyen d'un petit système de pompage pour l'irrigation dont la pression estimative était de 90 livres par pouce carré.

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1.0 INTRODUCTION

An adequate supply of water with quality that meets the needs for domestic consumption and other uses has always been necessary for success and growth of urban centres, industrial development, and agriculture. It has usually been considered that in Canada, and especially provinces such as Manitoba, there is generally an abundance of fresh water with good quality that can meet environmental and human development needs. Even so, increasing concerns regarding potential pollution of water resources in Manitoba as well as throughout the world has advanced demands by the public for better protection of water as more knowledge is gained on polluting sources and on the difficulties with combating pollution once it has occurred. There is further recognition that water quality and quantity cannot be taken for granted as increased demands from all sectors will continue to put greater pressures on available supplies, especially in the more heavily developed southern part of Manitoba. With the natural movement of water from one location to another, it is also understood that upstream accumulative effects can affect receiving downstream areas significantly. More recent changes in agriculture towards larger animal housing operations along with traditional practices of unencumbered livestock access to surface waters has helped trigger increased public concerns about environmental sustainable issues for this sector. Concerns are generally focussed on whether nutrients and pathogens in livestock manure, especially from larger, more densely populated operations, can be adequately managed to prevent excessive loss and unacceptable impacts to surface or ground waters.

Beneficial Management Practices (BMPs) is one of the primary mechanisms promoted by governments, the agricultural industry, and non-governmental organizations to help agriculture deal with mitigating potential contamination issues. While it is intuitively thought that incorporating BMPs into an operation will help reduce environmental impacts, there has been limited scientific study to determine their effectiveness, especially in areas such as Manitoba. As well, producers are hesitant to spend their limited resources on establishing and maintaining BMPs that may not provide any perceivable or measurable benefits. The Watershed Evaluation of Beneficial Management Practices (WEBs) Project was established to gather some of this information on field and associated small watershed scales. Traditional small-plot designs are often used for this type of scientific investigation because they can provide greater control of confounding factors that may affect treatments under study than larger scaled studies. However, transference of information from small plots to watershed scales may also not be completely successful because of difficulties to account for the myriad of interactions that generally occurs in the much larger natural setting. The WEBs Project attempted to better understand how BMPs would perform in natural field and small watershed scale situations given the multitude of interacting factors that usually occur.

The WEBs Project was initiated by Agriculture and Agri-Food Canada under the Greencover Canada program with Ducks Unlimited Canada being a key partner. It is a multidisciplinary project relying on expertise from different provincial and federal departments, universities, and other non-governmental groups to gather and assess data. From this information, the effectiveness and cost/benefits for certain BMPs are anticipated so assistance can be given to producers with making the most cost-effective, efficient choices to manage nutrients, pathogens, or other contaminants that could originate from their operations.

Escherichia coli (*E. coli*) is a member of a larger group of microbiological organisms known as fecal coliforms. Fecal coliforms have been one of the most widely accepted

microbiological organisms used as an indicator of pollution from sources of human or animal excrement. In recent years, the Manitoba Water Quality Management Section began using *E. coli* as the sole main indicator of fecal contamination in surface waters for a couple of reasons. First, *E. coli* is considered a more definitive indicator than the general fecal coliform test for originating from fecal matter of warm-blooded animals that includes humans, wildlife, waterfowl, and livestock. Unlike testing for *E. coli*, enumeration for the broader group of fecal coliforms may also indicate the presence of *Klebsiella*, a species that is not necessarily restricted to fecal sources. *Klebsiella* has been known to survive and replicate in organic rich environments such as waters receiving pulp and paper effluents (Bell *et al.* 1978, Huntley *et al.* 1976) so in certain environments may provide overestimates of fecal contamination.

The Stepler Watershed in the larger South Tobacco Creek Watershed was selected for the WEBs project because of several positive attributes. First, the family that owned and managed the land in this small watershed were willing participants. They have a keen interest in the relationship between agricultural practices and environmental issues that resulted in a positive history of helpful collaboration for past studies conducted in the South Tobacco Creek Watershed. Second, it was through these projects that a considerable amount of data such as land use, cropping, fertilization practices, and other information had been collected from the Stepler Watershed and surrounding lands within the South Tobacco Creek Watershed. Third, the Stepler Watershed appeared to have multiple characteristics that allowed evaluation of a variety of BMPs, yet was small enough to allow adequate management of day-to-day data collection needs. Additionally, infrastructure such as monitoring weirs and water sampling equipment was already established at one of the BMP sites, the Twin Watershed. In conjunction with operation of this equipment was added value from an experienced field technician who was familiar with producers and landscapes in both the Stepler Watershed and companion watersheds in the larger South Tobacco Watershed.

The main objectives for the *E. coli* component of the Stepler WEBs project were to determine:

1. the extent receiving surface waters in the watershed were affected by runoff contaminated from livestock manure, and
2. the effectiveness of a holding pond at capturing and holding runoff from a confined livestock holding area for a period of time to reduce transport of *E. coli* bacteria to downstream areas.

Secondary objectives included assessing:

- (i) *Escherichia coli* count reduction with time in the holding pond, and
- (ii) if *E. coli* counts declined as they passed through a small pump when holding pond water was used for irrigation.

The purpose of this report is to present results of the *E. coli* data collected for the WEBs Project in the Stepler Watershed, Manitoba, Canada, from 2004 to 2007.

2.0 DESCRIPTION OF STUDY AREA

2.1 General

The Stepler Watershed is located in southern Manitoba, Canada at approximately 98° 20' longitude; 49° 21' latitude (Figure 1). It is at the headwaters of the larger South Tobacco Creek Watershed. The Stepler Watershed and other headwater tributaries of South Tobacco Creek originate along the upper plateau of the Manitoba Escarpment. While elevations of the South Tobacco Creek within the escarpment are relatively steep and decline an average of approximately 2.14 m per 100 m distance, water courses in the Stepler Watershed have less gradient. Within the Stepler Watershed the gradient declines on average about 1.86 m for every 100 m. Water courses in the Stepler Watershed are also intermittent, usually flowing only during spring melt or after reasonably good precipitation events.

Flow from the Stepler Watershed and other tributaries to the South Tobacco Creek eventually form a relatively moderate gradient river between the base of the Manitoba Escarpment and the village of Miami, Manitoba declining approximately 0.8 m per 100 m in distance. The South Tobacco Creek eventually flows into the Red River via the Morris River where the gradient is considered fairly flat (average of about 0.032 m drop per 100 m distance). The South Tobacco Creek confluence with the Morris River occurs just north of the Town of Morris.

Soils in the Stepler Watershed and larger Tobacco Creek Watershed are generally Orthic Dark Gray Chernozem and Gleyed Dark Gray Chernozem with characteristics that are productive soils for crop production, fairly high water holding capacity, moderate infiltration and limited risk of groundwater impacts from leaching (Michalyna *et al.* 1988, Langman 1986). Surface deposits are characteristic of glacial origins with deep basin deposits of silt, sands, and clays in the lowlands (Weir 1983). Soils transition to coarser near shore sands and gravels on the escarpment upslope. Tills in the escarpment are moderately calcareous primarily derived from Mesozoic shales (Weir 1983).

Mixed broadleaf forest is predominant along riparian corridors of tributary and main river channels within the escarpment. Broadleaf species mainly consist of elm (*Ulmus americana*), burr oak (*Quercus macrocarpa*), maple (*Acer negundo*), green ash (*Fraxinus pennsylvanica*), and aspen (*Populus tremuloides*). The plains on the upper plateau and below the Manitoba Escarpment in this region are categorized as wooded grassland (Weir 1983) that is conducive to land use being mainly cropland (Manitoba Land Resource Unit 1997).

Climate in the southern part of Manitoba containing the Stepler Watershed has been termed continental (Phillips 1990). Weir (1983) summarizes average annual precipitation to be approximately 533 mm. There are between approximately 110 to 120 frost-free days during the growing season with mean July and January temperatures of approximately 21° C and -17° C, respectively.

2.2 Stepler Watershed

The Stepler Watershed is elongated along its drainage path and covers an area of approximately 206 ha (Figure 2). The upper part of the watershed contains two intermittent water courses; a north channel that runs through field units F1 and F3, and a southern channel

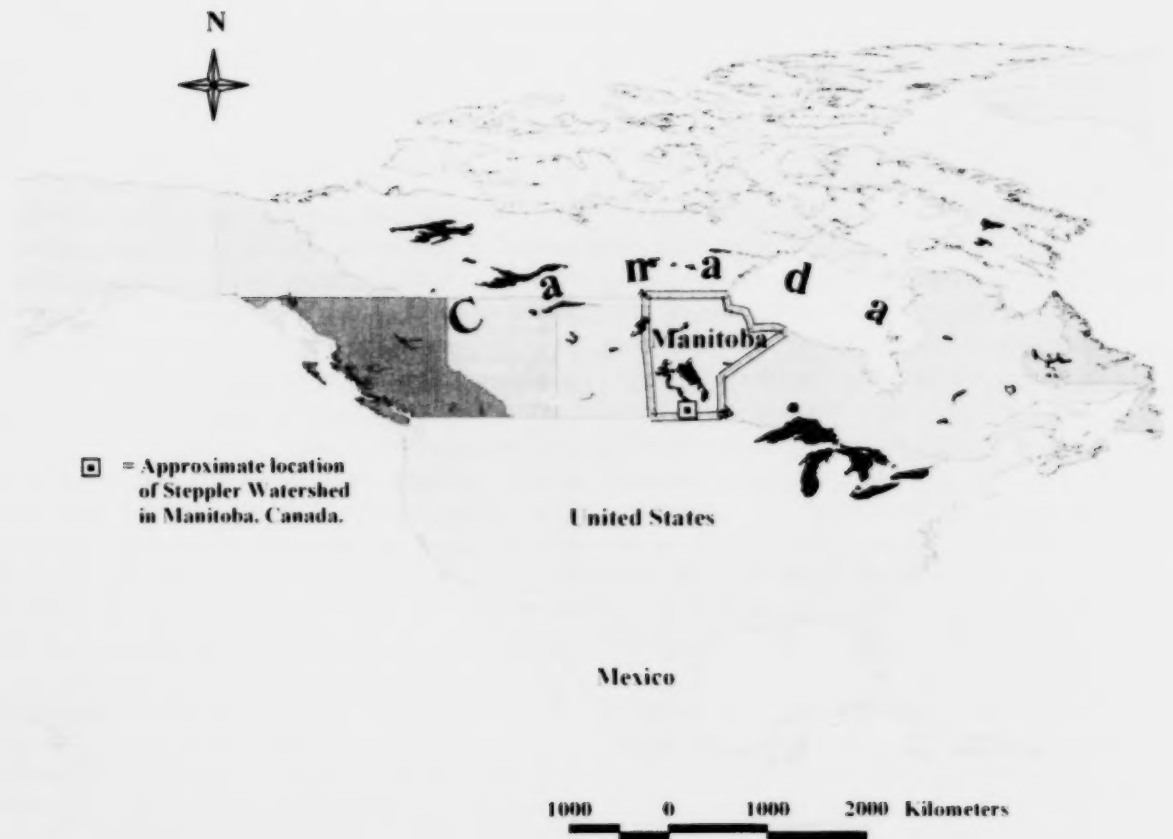


Figure 1. Geographic location of the WEBs Steppler Watershed Project, Manitoba, Canada, 2004 to 2007.

that runs through field units F2 and F4 (Figure 2). These two water courses join near the mid point of the watershed in field unit F5 to form a single main channel just downstream of sampling site MS5. Sampling site MS5 captures runoff from a livestock confinement area in the farmyard that is referred to in this report as the farmyard-cattle-corral (in watershed unit F6, Figure 2). This main channel eventually enters a small man-made reservoir that is at the outlet of the watershed (sampling site MS12). This reservoir was created by a small, earthen dam built in 1986 to help attenuate runoff flows and reduce channel erosion during heavy runoff from spring melt or precipitation events. The dam at the outlet is commonly called the Steppler Small Dam.

Prior to the main channel entering the small reservoir, two small intermittent drainage channels from field units F7 and F9 enter it. Water quality monitoring sites MS6 and MS8 were located at the outlets of field units F7 and F9, respectively. Sampling site MS9 was downstream of riparian improvements on field unit F8 (Figure 2).

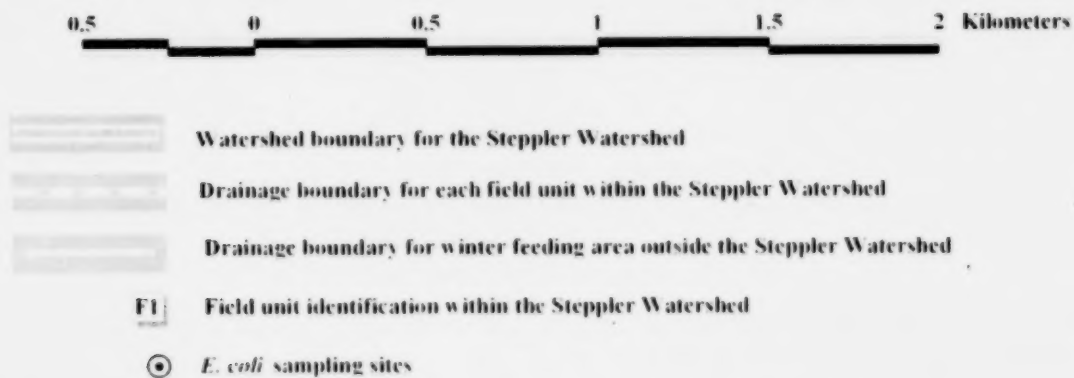
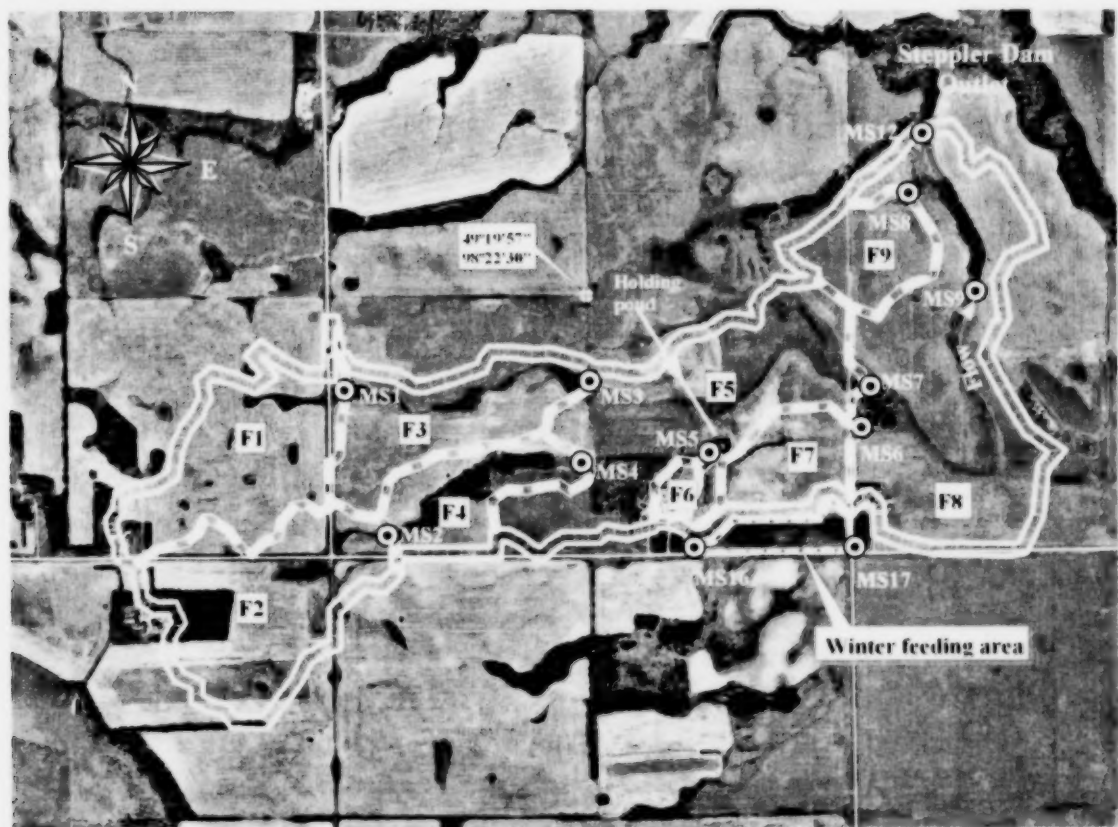


Figure 2. *Escherichia coli* sampling sites, individual field unit drainage boundaries, and boundary of the Steppler Watershed for the WEBs Steppler Watershed Project, Manitoba, Canada, 2004 to 2007.

Study design within the Stepler Watershed was based upon determining individual drainage areas for each field unit used for a treatment and establishing upstream and downstream water collection sites at key locations. Study fields and associated water sampling sites for *E. coli* measurements were as shown in Table 1, Figure 2.

Table 1. Field identification, land use descriptions, and treatments for field units in the WEBs Stepler Watershed Project, 2004 to 2007.

Identification	Field		Upstream water sampling site	Downstream water sampling site
	Use	Treatment		
F1	Annual cropland	Remains this way	-	MS1
F2	Annual cropland	Remains this way	-	MS2
F3	Annual cropland	Remains this way	MS1	MS3
F4	Annual cropland	Converted to forage	MS2	MS4
F5	Riparian and pasture areas in forage with limited control on cattle access	Converted to increased controlled grazing	MS3 & MS4	MS7
F6	Farmyard site with cattle corral	Runoff captured into a holding pond	-	MS5
F7	Annual cropland	Converted to forage	-	MS6
F8	Annual cropland	Remains in cropland with riparian area improved to enable harvesting of forage	MS6 & MS7	MS9
F9	Annual cropland	Remains this way	-	MS8

2.3 Background Sites

Escherichia coli data were periodically collected from three sites located outside the Stepler Watershed but within the larger South Tobacco Creek Watershed to provide some background information. One of the sampling sites (MS15) is at the outlet of a natural wooded area on legal description NE12-5-8WPM (section, township, range of west prime meridian) at approximately 49°22'46"; 98°25'12" (Figure 3). This watershed unit is approximately 20.2 ha (50 acres) and is comprised entirely of natural bush. It is approximately six kilometres northeast of the Stepler Watershed.

The other two background sites were in the Madill Watershed. The Madill Watershed is

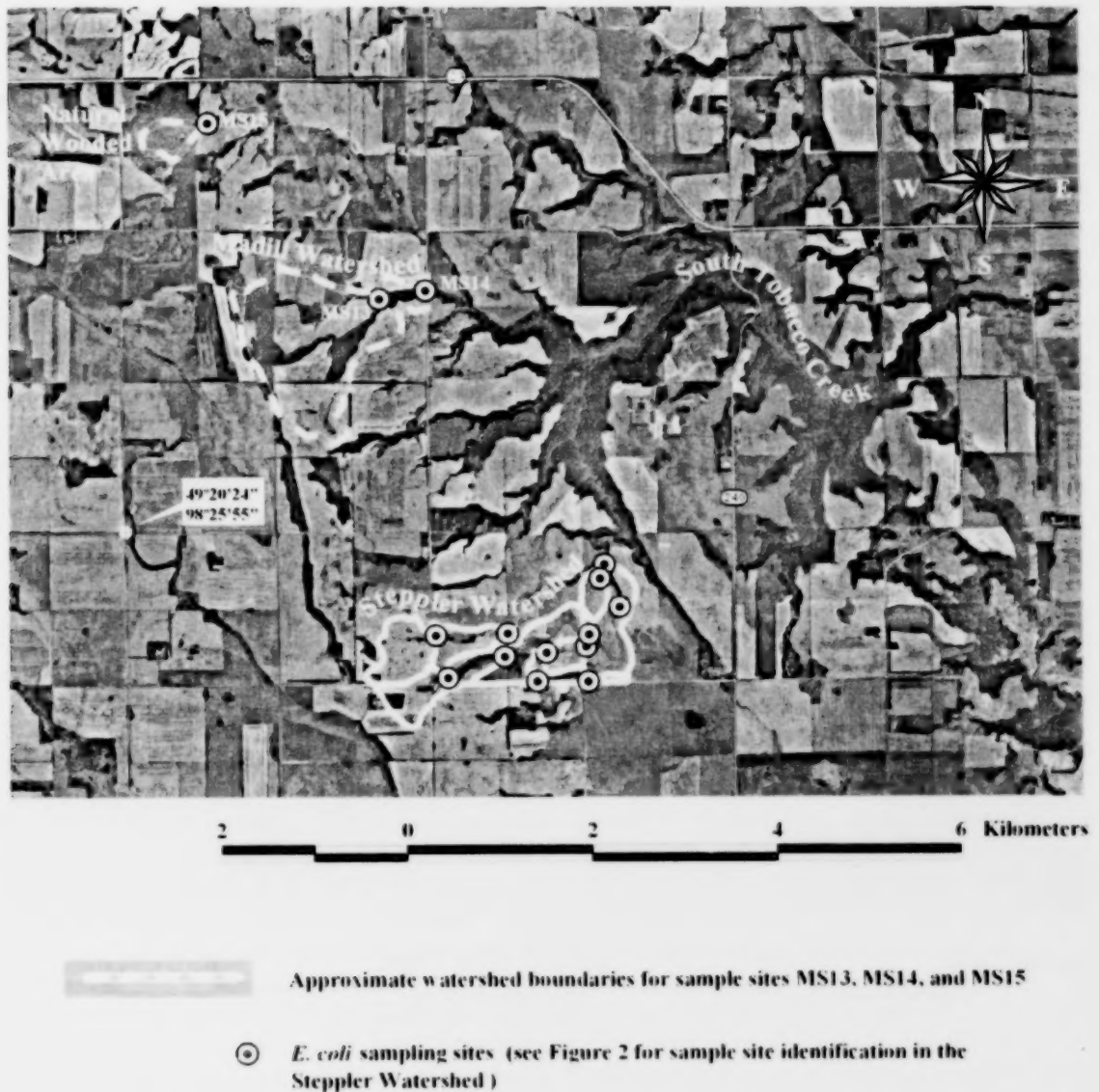


Figure 3. Location of sampling sites MS13, MS14, and MS15 that provided background *Escherichia coli* counts in the WEBs Stepler Watershed Project, Manitoba, Canada, 2004 to 2007.

located approximately two kilometres northeast of the Stepler Watershed. Similar to the Stepler Watershed, a small dam (Madill Small Dam) was constructed at the outlet of this watershed in 1988 to help reduce soil erosion and channel scouring during heavy runoff events. The sampling site (MS13) upstream of the reservoir in this watershed is located on legal description NW31-4-7WPM at approximately 49°21'17"; 98°22'22". Site MS14 is at the outlet of the small dam in the Madill Watershed on legal description NE6-5-7WPM at

approximately 49°21'17"; 98°22'22" (Figure 3). The drainage area for the Madill Watershed is approximately 220 ha (545 acres).

2.4 Groundwater Test-wells

During 2006 and 2007 *E. coli* samples were collected from four shallow test-wells located along the drainage route from the farmyard-cattle-corral to the inlet of the holding pond (MS5) to help determine if groundwater subsurface flow was contributing to holding pond volumes. Well depths were approximately 0.5 m below the drainage channel bottom elevation. These test-wells were DTW1, DTW2, DTW3, and DTW4 (Figure 4).

In 2007, an *E. coli* sample was also collected from each of two deeper borehole test-wells closer to the holding pond to determine if there was any evidence that bacterial contamination was moving deeper than the shallow subsurface test-wells. Well depths were approximately 3.5 m from surface. The borehole test-wells were BH9 and BH11 (Figure 4).

3.0 METHODS

3.1 General

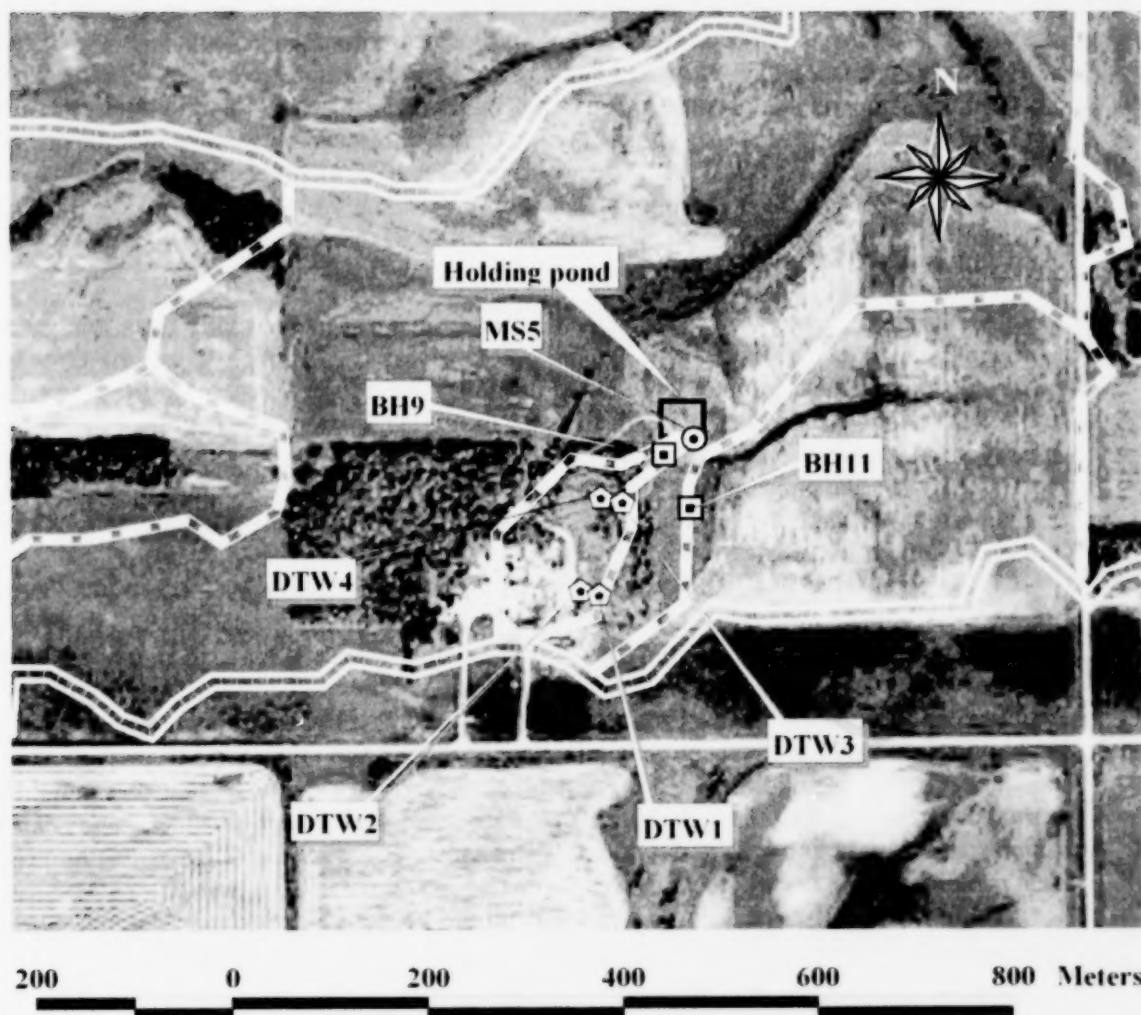
The first two years (2004 and 2005) of the four-year WEBs project provided background conditions prior to establishment of the BMPs being studied. Post BMP implementation was provided by the 2006 and 2007 data. *Escherichia coli* samples were collected from all water sampling sites (MS1 to MS9 and MS12) within the Stepler Watershed during 2004. The 2004 data was then evaluated to determine the extent of *E. coli* contamination in the watershed. This assessment determined where reductions in sampling for *E. coli* could occur and where it needed to be maintained for 2005 to 2007 to remain within study resources and to not jeopardize study objectives.

Escherichia coli bacteria were used as an indicator organism for the potential presence or absence of other more harmful pathogens that is a common practice in many jurisdictions. Higher numbers of *E. coli* are generally considered to indicate there is a greater probability that higher numbers of more harmful pathogens also exist and vice versa. However, it must be understood that the presence of *E. coli* does not necessarily mean an equal presence of more harmful pathogens nor does the absence mean more harmful pathogens were absent or in low numbers.

3.2 Field Collections

Escherichia coli samples were collected as discrete grab water samples from each of the water sampling sites. Each *E. coli* water sample was collected into a 125 mL bottle supplied by ALS Laboratories¹ and all samples were submitted to the commercial laboratory within 24

¹ ALS Laboratories, Unit 12, 1329 Niakwa Road East, Winnipeg MB, R2J 3T4.



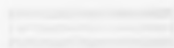
-  Watershed boundary for the Stepler Watershed
-  Approximate feld unit drainage boundary within the Stepler Watershed
-  *E. coli* sampling sites
-  Farmyard-cattle-corral drainage channel test-well approximate location
-  Borehole test-well approximate location

Figure 4. *Escherichia coli* sampling sites for shallow test-wells near the farmyard-cattle-corral drainage channel and holding pond borehole test-wells in the Stepler Watershed Project, Manitoba, Canada, 2004 to 2007.

hours of collection. On days when *E. coli* samples were collected, only one sample from a site usually was taken. On occasion, additional *E. coli* samples from some sites were collected at a different time interval on the same day, especially those associated with the holding pond. Collections of *E. coli* samples often coincided with gathering water samples from automatic sampling devices that were established to capture water samples for nutrient and other chemical analyses.

3.3 Laboratory Analyses

Escherichia coli in water samples collected from most sites usually were analyzed by the membrane filter technique (MF). Sample volumes of 10, 1, and 0.01 mL dilutions were extracted from the field samples and poured through a membrane filter. The filter was placed into a Petri Dish prepared with m-Coli Blue 24 broth. The inverted plates were incubated at 35° C for 24 hours. Colonies on the filter that turned blue were counted as *E. coli* due to reaction between the enzyme beta glucuronidase and BCIG (5-bromo-4 chloro-3 indolyl-beta-D-glucuronide) while other coliforms that reduced TTC (2,3,5 triphenyltetrazolium chloride) in the medium turned red.

Exceptions to use of the MF technique occurred for highly turbid water samples originating from the farmyard-cattle-corral that entered the holding pond (MS5 – inlet to the holding pond) and water from within the holding and that was used for irrigation (sites MS5A, MS5B, MS5C, and MS5D). These samples were enumerated by a 3-tube fermentation technique and expressed as most-probable-number (MPN). The MPN method was used for the highly turbid waters because it has a longer recovery period than the MF method. It was thought the MPN method would provide better recovery of organisms that may have been injured or stressed due to the turbidity. It was also considered that particulate matter in these samples could mask some bacterial colonies and create potential interferences to the m-Coli Blue medium used in the MF technique.

Both methodologies followed general procedures from Standard Methods for the Examination of Water and Wastewater (2005).

3.4 Statistical Analyses

Systat v11.0 statistical program was used for the statistical procedures. Prior to performing statistical analyses, all *E. coli* data were $\log_{10}$ transformed to increase normality from often naturally high variability in *E. coli* counts as well as to help compensate for variability introduced from the two laboratory analytical methodologies. Statistical differences ($\alpha = 0.05$) in comparisons of mean values between sites and for comparisons between years were determined by Analyses of Variance (ANOVA). The Bonferroni post-hoc test was used when comparison of means between sites was less than five pairs that included comparisons of between-years for individual sites. The Tukey post-hoc test was used where there were five or more mean pairs and included comparison of means between sampling sites.

Least squares linear regression analysis was used to help assess the number of days it took for *E. coli* within the holding pond to die-off and reach 200 *E. coli* 100 mL⁻¹ or less. This target value was chosen because it is the guideline limit in Manitoba for primary recreational activities (Williamson 2002). Primary recreational activities include those actions where human contact with water is high such as swimming. First, regression analyses was

performed in each year on holding pond *E. coli* counts that represented the start of a relatively steady decline after the initial, main runoff period ended. The approximate end of the main runoff period was determined from annual hydrographs. The number of days between the start of runoff to when the initial runoff was considered to have ended was simply added to regression estimates to determine the total number of days from the start of runoff to when *E. coli* counts declined to 200 *E. coli* 100 mL⁻¹.

Censored data reported at laboratory detection limits (e.g., < 3 *E. coli* 100 mL⁻¹) were assumed to be at that value (i.e., 3 *E. coli* 100 mL⁻¹) for calculation purposes. For less than detection limits this was a more conservative approach and provided slightly higher calculated values than if the actual concentration was known. However, an upper detection limit of >110,000 *E. coli* 100 mL⁻¹ was usually used between 2004 and 2006. Calculations based upon assuming *E. coli* counts at this upper limit would provide lower calculated values than if actual *E. coli* counts had been determined.

4.0 RESULTS AND DISCUSSION

4.1 General

The majority of *E. coli* samples between 2004 and 2007 were collected during the spring runoff period (Appendices 1 to 5). Spring runoff generally began in the last week of March and data up to the end of May was included as spring runoff for this *E. coli* assessment. The greatest recharge period for rivers and lakes in southern Manitoba usually occurs during spring runoff. Runoff in the Steppler Watershed from 2004 to 2007 generally followed this pattern (Appendix 6). Spring runoff volumes are generated from melt-water of snow that accumulates during a sub-zero degree Celcius ($< 0\text{ }^{\circ}\text{C}$) period usually extending from approximately mid-November to late March or early April. Additional runoff often occurs from spring-time rainfall events. As indicated from hydrographs shown in Appendix 6, precipitation and subsequent runoff events that generated flow in the main drainage channels were usually limited after June with only an occasional *E. coli* sample collected between July to October (Appendices 5f, 5h, 5i, and 5j). The exception to this pattern occurred in 2005 when there were large enough rainfall events in July that produced substantive runoff events (Appendices 6a to 6d).

4.2 2004 Preliminary Assessment

During the first year of the WEBs study (2004), *E. coli* samples were collected at all water quality sampling stations within the Steppler Watershed (MS1 to MS9 and MS12) plus the three background sites MS13, MS14, and MS15 that were in other watersheds within the larger South Tobacco Creek Watershed (Figures 2 and 3). These data were evaluated to determine how much *E. coli* contamination was occurring throughout the watershed area, and whether sampling could be discontinued at some sites while still allowing adequate assessment of contributions coming from the farmyard-cattle-corral (site MS5). Mean *E. coli* counts during spring runoff in 2004 indicated relatively low counts at sites upstream of MS5 (sites MS1 to MS4) or from the background sites MS13, MS14, and MS15. Mean *E. coli* counts at sites MS1 to MS4 ranged from 20 *E. coli* 100 mL⁻¹ to 51 *E. coli* 100 mL⁻¹ compared to a mean count of 13,836 *E. coli* 100 mL⁻¹ from the farmyard-cattle-corral runoff site MS5 (Figure 5). Pairwise comparisons of the means also indicated that there were no significant differences ($\alpha=0.05$) between mean values for all sites except for MS5 (Appendix 7). *Escherichia coli* counts from site MS5 were significantly different than all sites except MS6.

Based upon the 2004 data, it was considered that maintaining the upstream site MS4 and the downstream sites MS7, MS9, and MS12 would provide a reasonable data set to assess how contributions from the farmyard-cattle-corral (site MS5) may be impacting upon downstream areas. Sampling from sites MS1, MS2, MS3, MS6, and MS8 were not continued on a regular basis for the following reasons. First, *E. coli* counts at sites MS1, MS2, and MS3 were fairly low and well under 200 *E. coli* 100 mL⁻¹, the water quality guideline for protection of primary recreational activities in Manitoba. Sites MS1 and MS2 were also upstream of sites MS3 and MS4 (Figure 2). Site MS3 was at the outlet of field F3 that was in annual cropland and it was intended to stay this way. Meanwhile, site MS4 was downstream of field F4 that was being converted to forage so it was considered this field and sampling site would provide

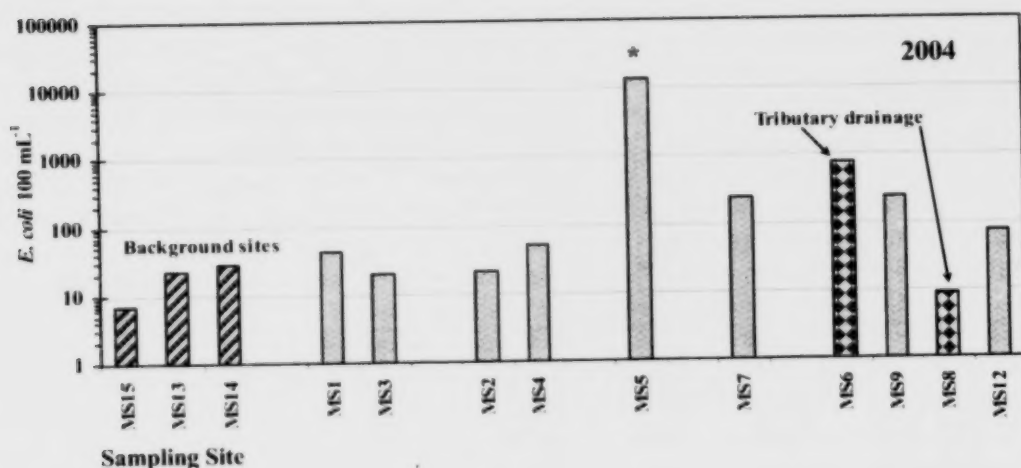


Figure 5. Mean *Escherichia coli* counts (*E. coli* 100 mL⁻¹) from sites in the WEBs Steppler Watershed Project, 2004.

Note: * = mean value was significantly different ($p < 0.001$) from mean values at other sites, except for MS6.

- Sites MS1 to MS12 arranged in approximate upstream to downstream order (see Figure 2).
- y-axis is on a logarithmic scale.

the best upstream conditions to compare with sites downstream of MS5. Mean *E. coli* counts from MS4 in 2004 were also greater than from MS3 (Figure 5).

Sites MS6 and MS8 were not maintained on a regular basis because they were not on the main drainage channel between sites MS7 and MS12. They were outlets for the smaller, intermittent, tributary drainage from field units F7 and F9. The mean *E. coli* count at site MS8 was also lower than at upstream sites MS1 to MS4 and background sites MS13 and MS14 (Figure 5). As well, MS8 represented drainage from field F9 that was in annual crop (Table 1, Figure 2). It was not expected that *E. coli* counts would significantly change in subsequent years because the land use for field F9 was staying the same and previous studies found significantly less fecal coliform bacteria counts in areas without cattle compared to those grazed by cattle (Mankin *et al.* 2006, Bohn and Buckhouse 1981, Doran and Linn 1979).

The relatively high mean *E. coli* count from MS6 in 2004 was unexpected. Four samples collected from MS6 between March 25 and March 30, 2004 were the main contributors to the high mean value in 2004. These samples ranged from 1,330 to 4,300 *E. coli* 100 mL⁻¹ (Appendices 1 and 5g). Site MS6 was the downstream collection point for field F7 (Figure 2). Some detection of *E. coli* counts from F7 could be expected due to potential presence of small mammals such as rabbits (*Lepus spp.*), deer mice (*Peromyscus maniculatus*), or ground squirrels (*Citellus spp.*). Harms *et al.* (1975a and 1975b), Robbins *et al.* (1972), Kunkle (1970) among others found fecal coliform counts in non-grazed, non-manured fields could often exceed primary contact criteria for swimming (200 organisms 100 mL⁻¹) while Doran and Linn (1979) attributed incidences of higher bacteria counts in their non-grazed, control field to observed increases in wildlife activity. However, it had not been expected that *E. coli* counts at site MS6 would be elevated as much as observed in 2004.

because disturbance by annual cropping would disrupt habitat and keep non-transient rodent populations relatively low.

A review of field observations recorded during *E. coli* sample collections in 2004 revealed the elevated counts at site MS6 did not appear to originate from its source field F7. These high counts could be attributed to an unusual circumstance at that time due to an unperceived weakness with the physical characteristic of the channel draining the farmyard-cattle-corral. During the runoff period when the elevated *E. coli* observations occurred, some runoff from the farmyard-cattle-corral actually "short-circuited" from the channel draining this area and overflowed to field F7 instead of following its normal route to sampling site MS5. This overflow then migrated through the drainage path of field F7 to the outlet at site MS6. Elevations separating drainage divides between field units in the Stepler Watershed were not always very large. During 2004, it was also observed that part of the drainage path between the farmyard-cattle-corral and site MS5 had been filled with a considerable amount of snow. This snow-pack restricted free passage of the spring-melt along its natural course. As spring-melt was often quicker in the more disturbed and exposed cattle corral area, this caused some back-flooding upstream of site MS5 and further assisted with overflow to field F7. To alleviate this condition from happening again in subsequent years, moderate excavation was done on the drainage route to MS5 to enhance the channel capacity. As well, prior to the commencement of spring runoff in the following years, snow was partially cleared from the channel and placed on the bank of the drainage route from the farmyard-cattle-corral to ensure free flow of runoff to site MS5. Re-occurrence of similar high *E. coli* counts at MS6 was not expected with these modifications. Field observations and the very limited sampling during spring runoff in 2005 and 2006 indicated the modifications were successful (Appendix 5g).

4.3 2004 to 2006 Stepler Watershed *E. coli* Counts

Mean *E. coli* counts during spring runoff from most sampling sites in the Stepler Watershed from 2004 to 2007 were usually within the water quality guideline for protection of primary recreational activities in Manitoba that is 200 *E. coli* 100 mL⁻¹ (Figure 6). The consistent exception where mean *E. coli* counts well exceeded 200 *E. coli* 100 mL⁻¹ of water was in runoff from the farmyard-cattle-corral (MS5). Mean spring values from site MS5 ranged from approximately 3,758 *E. coli* 100 mL⁻¹ to 23,442 *E. coli* 100 mL⁻¹ during 2007 and 2005, respectively (Figure 6, Table 2).

Daily *E. coli* counts from MS5 also were usually much higher than from other sampling locations in the Stepler Watershed. While an occasional high to very high *E. coli* count (i.e., in the 1,000s of *E. coli* 100 mL⁻¹ to greater than 100,000 *E. coli* 100 mL⁻¹) was observed at some other sampling sites, these elevated counts were considerably more common from MS5 (Appendices 1 to 5). During each year, the maximum *E. coli* count was also observed in runoff at MS5. The maximum count recorded from 2004 to 2007 was 2,300,000 *E. coli* 100 mL⁻¹ during 2007 (Table 3). *Escherichia coli* counts from the farmyard-cattle-corral as high or higher than observed in 2007 were anticipated since Brockamp (1993) reported fecal coliform counts in runoff originating from nearby cattle feedlots averaged about 9,000,000 100 mL⁻¹ to a maximum of approximately 53,000,000 100 mL⁻¹. Ramos *et al.* (2005) also found presumptive fecal coliforms in runoff from cattle manure slurry applications to a sandy loam to be as high as 1,100,000 100 mL⁻¹.

It is possible that the high values observed at MS5 during 2007 could have also

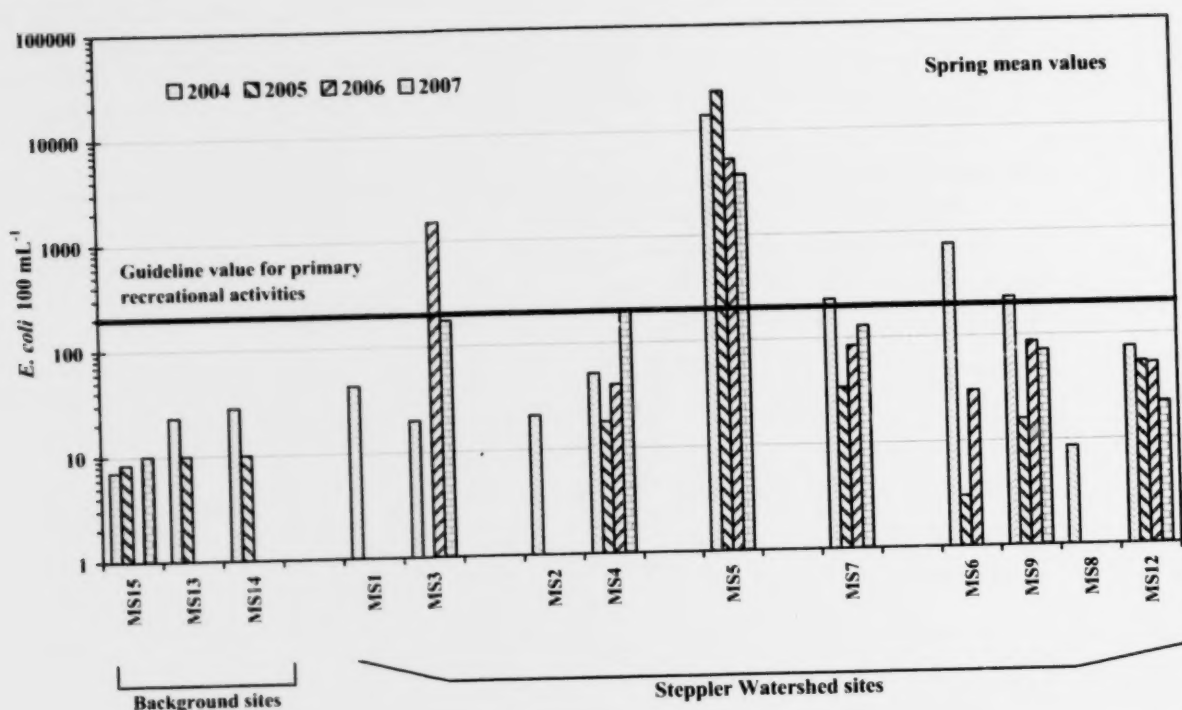


Figure 6. Mean *Escherichia coli* counts (*E. coli* 100 mL⁻¹) during spring runoff from the WEBS Stepler Watershed Project, 2004 to 2007. Sites MS1 to MS12 arranged in an upstream to downstream order. Spring runoff includes from the start of spring melt to the end of May.

occurred in 2004, 2005, or 2006. However, the upper laboratory detection limit was usually set at $>110,000$ *E. coli* 100 mL⁻¹ that precluded determining if or when higher counts occurred in these years. It was considered determining counts to this detection level was more than enough to indicate if substantial contributions were occurring to contaminate surface waters. One sample in each year of 2004 and 2005 from MS5 was analyzed to an endpoint enumeration of 430,000 *E. coli* 100 mL⁻¹. This showed substantially higher counts than the upper analytical limit of $>100,000$ *E. coli* 100 mL⁻¹ normally used in the laboratory analyses had occurred in these years. Therefore, it was possible that some of the other observations reported as $>110,000$ *E. coli* 100 mL⁻¹ in 2004 to 2006 may have matched or exceeded the maximum values found in 2007.

Figure 6 shows that the mean value from sampling site MS3 in 2006 was elevated compared to mean values from other years as well as from sites other than sampling site MS5 (farmyard-cattle-corral runoff). Although maximum *E. coli* counts at site MS3 between 2004 and 2007 were generally above the recreational guideline value of 200 *E. coli* 100 mL⁻¹, the high mean value observed during spring runoff 2006 was due to only a single sample collected on May 25 of that year (Table 3). Field observations also revealed this value coincided with

Table 2. Mean *Escherichia coli* counts (*E. coli* 100 mL⁻¹) during spring runoff from the WEBs Steppler Watershed, 2004 to 2007. Spring runoff includes data from the beginning of runoff to the end of May. Note: Mean values based upon log₁₀ transformed data and then converted back to non-logarithm values for presentation purposes.

Sampling Site Reference	Sampling Site ID	Year			
		2004	2005	2006	2007
		(E. coli 100 mL ⁻¹)			
Background Sites	MS15	7	8		10
	MS13	23	10		
	MS14	28	10		
Upstream for field F3	MS1	43			
Downstream for field F3	MS3	20		1,500	173
Upstream for field F4	MS2	21			
Downstream for field F4	MS4	52	18	40	195
Farmyard-cattle-corral	MS5	13,836	23,442	5,284	3,758
Downstream of MS3, MS4, and MS5	MS7	233	34	84	129
Downstream for field F7	MS6	753	3	30	
Downstream of MS7 and MS6	MS9	229	16	86	71
Downstream for field F9	MS8	9			
Steppler Watershed and Steppler Dam Outlet	MS12	74	53	52	22

cattle grazing just upstream in the nearby riparian area beginning on May 19, 2006 (Appendix 5d). It appeared that fresh contributions by cattle were being quickly transferred to the intermittent water courses when runoff occurred. Stephenson and Street (1978) found cattle presence directly affected fecal coliform densities in south-western Idaho streams with counts increasing rapidly after cattle were introduced to watersheds and remained high until cattle were removed. Jawson *et al.* (1982) also observed some correlation between recentness of grazing and amounts of indicator bacteria in runoff. Cattle put into a north-western Idaho watershed 3 d prior to a June runoff event in 1977 coincided with their highest observed fecal coliform, total coliform, and fecal streptococci counts. The elevated mean value from site MS3 in the Steppler Watershed during 2006 not only reinforced how results could be affected greatly by a very limited number of samples, but also indicated how important timing and

Table 3. Maximum *Escherichia coli* counts (*E. coli* 100 mL⁻¹) in annual runoff from the WEBs Stepler Watershed Project, 2004 to 2007. Sample numbers are in parenthesis.

Sites	Spring				Summer/Fall			
	2004	2005	2006	2007	2004	2005	2006	2007
MS15	93 (4)	< 10 (5)		10 (2)	99 (4)	20 (1)		100 (1)
MS13	4,300 (10)	< 10 (1)			930 (5)			
MS14	380 (10)	10 (1)			2,300 (4)			
MS1	1,200 (8)				23 (1)			
MS3	230 (12)	< 10 (1)	1,500 (1)	15,000 (15)	230 (5)			3,000 (6)
MS2	430 (11)				23 (1)			
MS4	930 (12)	2,300 (8)	110,000 (8)	16,000 (13)	1,200 (2)	2,300 (5)		1,440 (6)
MS5	430,000 (12)	>110,000 (13)	> 110,000 (14)	2,300,000 (24)	110,000 (5)	430,000 (8)		2,300,000 (17)
MS5A			> 110,000 (11)	93,000 (24)			2,300 (16)	93,000 (13)
MS5B			46,000 (8)					430,000 (8)
MS5C			430 (8)	2,300 (6)			430 (11)	1,500 (1)
MS5D			280 (6)				2,300 (8)	
MS7	47,000 (13)	1,270 (9)	2,900 (10)	54,000 (19)	4,300 (5)	9,300 (8)	230 (1)	11,900 (13)
MS6	4300 (6)	9 (2)	30 (1)		4,300 (1)			
MS9	9,300 (12)	70 (7)	2,700 (9)	290,000 (18)	2,300 (4)	560 (7)	50 (1)	4,000 (12)
MS8	150 (4)							
MS12	9,300 (11)	600 (8)	400 (10)	1,480 (15)	93 (2)	940 (6)	< 3 (1)	350 (8)

Note: Spring encompasses runoff data from start of spring melt to the end of May while summer/fall includes data from June 1 to freeze-up.

location for livestock management may be on the effectiveness of some BMPs.

The lowest mean *E. coli* counts between 2004 and 2007 came from the background site MS15 and ranged between 7 to 10 *E. coli* 100 mL⁻¹ (Figure 6). Daily *E. coli* counts from site MS15 were also relatively low with maximum values less than 100 *E. coli* 100 mL⁻¹ (Table 3, Appendix 5a). Low *E. coli* counts from site MS15 were generally expected since it was at the outlet of a small watershed that was naturally wooded and where the only contributions would be from wildlife inhabiting the area. Niemi and Niemi (1991) found fecal bacteria indicators were usually detected in only about half the samples from pristine areas compared to those from agricultural watersheds. They also found maximum counts from pristine areas were less than median values of agricultural watersheds and attributed them to wildlife such as elk (*Alces alces*) and white tailed deer (*Odocoileus virginianus*) living within those areas.

Mean *E. coli* counts from the other two background sites MS13 and MS14 (Madill Watershed, Figure 3) were also relatively low and well under the guideline for primary recreational activities (Figure 6). Although limited sampling occurred at these sites, daily samples from sites MS13 and MS14 during 2004 showed a greater variation and higher *E. coli* counts than observed at site MS15. Maximum daily counts at site MS13 during spring runoff and site MS14 in summer runoff were 4,300 *E. coli* 100 mL⁻¹ and 2,300 *E. coli* 100 mL⁻¹, respectively (Table 3, Appendix 5b). The exact cause for these elevated values at the two sampling sites is not known, but wildlife such as beaver (*Castor canadensis*) can inhabit the pond at the Madill Dam. Cattle presence and larger wildlife such as whitetail deer can also occur on open forage and riparian areas in the Madill Watershed. Therefore, opportunities for occasionally elevated *E. coli* counts exist.

4.3.1 Holding Pond BMP Assessment

The effectiveness of the holding pond on *E. coli* counts downstream of contributions from the farmyard-cattle-corral was not readily evident or fully understood based upon data collected from 2004 to 2007. During 2004 and 2005 when the holding pond was not in operation, mean *E. coli* counts at MS7 (first site downstream of runoff from MS5) were greater than found at the upstream site MS4 (Figure 6). This indicated runoff from the farmyard-cattle-corral (MS5) was increasing counts downstream as could be expected. *Escherichia coli* counts at MS5 usually were significantly greater ($\alpha = 0.05$) than counts at the other sites during all years that also indicated contributions from the farmyard-cattle-corral had potential to significantly impact upon the downstream site MS7 (Appendix 7). An exception occurred during 2007 where the mean *E. coli* count from MS5 was not significantly different ($\alpha = 0.05$) but still considerably higher than the mean count at upstream site MS4 (Figure 6).

Under operation of the holding pond during 2006 and 2007, the opposite of what was observed in 2004 and 2005 was anticipated. *Escherichia coli* counts at site MS7 downstream of runoff from the farmyard-cattle-corral (MS5) were expected to be the same and possibly lower than what was found at the upstream sites MS4 or MS3. With the holding pond intercepting and holding highly contaminated runoff from the cattle-corral-area, counts at MS7 could be reduced through die-off from exposure to the sunlight's ultraviolet radiation or be diluted by uncontaminated runoff occurring between these sites. A lower mean *E. coli* count at the downstream site MS7 compared to the upstream sites MS3 or MS4 during 2007 indicated the holding pond was intercepting and reducing transport of *E. coli* contributions as

expected (Figure 6). However, effectiveness of the holding pond remained unclear because during spring runoff 2006 the mean *E. coli* count at site MS7 was slightly higher than the mean count from the upstream site MS4 (Figure 6). Although the mean count at MS7 was lower than at the other upstream site MS3, the MS3 mean value in 2006 was represented by only one sample so it was not considered representative of overall spring conditions nor suitable for this comparison. Additionally, there were no significant differences ($\alpha = 0.05$) between *E. coli* counts at MS3 or MS4 to the downstream site MS7 for any year from 2004 to 2007 (Appendix 7). Even between-year mean *E. coli* counts from 2004 to 2007 for each site MS4, MS5, or MS7 showed there were no statistical differences ($\alpha = 0.05$). This indicated *E. coli* counts from the farmyard-cattle-corral were not affecting counts at the downstream site MS7 any more significantly before or after operation of the holding pond. It appeared the elevated *E. coli* counts in runoff from the farmyard-cattle-corral were being substantially reduced by natural attenuation through dilution from other downstream runoff or die-off by the time sampling site MS7 was reached. Contributions from other sources between the downstream site MS7 and upstream sites also appeared to be a confounding issue.

A factor that was initially considered to have potentially increased *E. coli* counts at MS7 during the early stages of spring runoff in 2006 was premature discharge from the holding pond emergency overflow spillway. In 2006 it only took about a week (March 31 to April 6) to fill the holding pond to capacity. Once the holding pond was filled to capacity, excess runoff exited out an emergency overflow spillway that was part of the design to help prevent damage to the top of the dykes. This discharge shares the same natural drainage route through field F5 to sampling site MS7 that runoff took prior to the holding pond being built (Figure 2). However, even though *E. coli* counts in the spillway runoff (site MS5B) were between 46,000 *E. coli* 100 mL⁻¹ on April 10 to 1,500 *E. coli* 100 mL⁻¹ on April 19, 2006, these elevated values did not show up at the downstream site MS7 during this period. Surprisingly, *E. coli* counts at site MS7 only ranged between 50 *E. coli* 100 mL⁻¹ to 80 *E. coli* 100 mL⁻¹ and stayed relatively low until May 25 (Appendices 5f and 5h). It appeared the relatively high *E. coli* counts from the emergency overflow discharge of the holding pond were somehow being ameliorated and had not significantly affected counts downstream at site MS7.

The distance between the overflow discharge of site MS5 and drainage path to site MS7 was over 500 m. At the Steppler Watershed, this distance appeared great enough to reduce *E. coli* counts either by dilution from other uncontaminated runoff or by providing enough ultraviolet light exposure time downstream of MS5 to cause significant die-off during 2006. *Escherichia coli* strains have shown survival times in river and well water from 1 to 7 d with greatest survival at 4° C (Personné *et al.* 1998, Ozkanca and Flint 1997) so it seemed probable that dilution in the Steppler Watershed may be a more significant factor. However, a certain amount of increased bacteria exposure to sunlight may have also occurred. Runoff flows in drainage channels this early in the thawing period in Manitoba can be slowed down by unmelted ice and snow blockage or by variable weather causing drops in temperature below freezing. Water depths in drainage channel runoff are also shallow helping to increase bacteria exposure to sunlight.

Although the distance between sampling sites MS5 and MS7 may have helped with *E. coli* count reductions in early spring of 2006, the much larger distance between the upstream sites MS3 and MS4 to downstream at MS7 also appeared to provide opportunities for additional *E. coli* sources between these sites that was another confounding factor. The distance between sites MS3 and MS4 to site MS7 was in the range of approximately 1000 m (about double the distance between farmyard-cattle-corral site MS5 and downstream to MS7).

This allowed potential contributions from additional sources that included small mammals such as field mice, ground squirrels, rabbits, and other animals inhabiting riparian and grassland areas between these sites. Moreover, timing of cattle presence in pasture and riparian areas, especially during 2006 and 2007, revealed this was a major influence affecting bacteria counts at sampling sites that helped obscure understanding the effectiveness of the holding pond on downstream counts.

A comparison between daily *E. coli* counts from sites MS4 and MS7 and field observations taken during sample collections showed elevated *E. coli* counts tended to coincide with the nearby presence of cattle. For example, at site MS4 in 2006 daily counts from start of runoff up to runoff events on May 11 were low and only ranged between < 10 *E. coli* 100 mL⁻¹ to 30 *E. coli* 100 mL⁻¹ (Appendix 5e). On May 19, cattle were moved into the riparian areas of fields F3, F4, and F5. These fields included areas immediately upstream of sampling sites MS3, MS4, and MS7 (Figure 2). Subsequent samples collected from MS4 and MS7 during a runoff event on May 25, 2006 were 110,000 *E. coli* 100 mL⁻¹ and 2,900 *E. coli* 100 mL⁻¹, respectively (Appendices 5e and 5h). Similarly, at sites MS3 and MS4 in 2007, daily *E. coli* counts were usually less than 100 *E. coli* 100 mL⁻¹ for runoff samples from early April. Meanwhile, water samples collected from these sites between May 22 and 23, 2007 resulted in bacteria counts ranging from 2,900 *E. coli* 100 mL⁻¹ to 16,000 *E. coli* 100 mL⁻¹ (Appendices 5d and 5e). These elevated *E. coli* counts corresponded to cattle having been moved into the riparian areas just upstream of MS3 and MS4 on May 16, 2007 and kept in these areas until they were moved out again on May 26, 2007. Subsequent runoff samples between May 27 and May 31, 2007 after the cattle had been removed revealed greatly reduced and relatively low *E. coli* counts that only ranged from approximately 67 *E. coli* 100 mL⁻¹ to 800 *E. coli* 100 mL⁻¹ (Appendices 5d and 5e). While relatively significant reductions of *E. coli* in runoff were almost immediate with removal of cattle from an area in the Steppler Watershed, bacterial counts were not as responsive in studies elsewhere. Fecal coliform bacteria could still exceed 200 organisms 100 mL⁻¹ more than a year after cattle were removed from a watershed in Idaho (Jawson *et al.* 1982) or 100 d after simulated rainfall events occurred in a study monitoring effects on cowpies (Kress and Gifford 1984). Small-plot runoff studies in Nebraska by Thurston-Enriquez *et al.* (2005) also demonstrated that fresh cattle manure could act as a potential long-term source of microbial loads including *E. coli*.

Despite uncertainty on how effective the holding pond mitigated significant transport of *E. coli* counts to downstream areas any more than natural attenuation, the study showed that the holding pond was effective at capturing runoff and substantially reducing these counts prior to release of water from the pond. Counts of *E. coli* entering the holding pond were often in the range of 10^4 to 20^5 *E. coli* 100 mL⁻¹ while counts were orders of magnitude lower as the pond was emptied. A variety of factors such as livestock presence at sites, frequency and magnitude of runoff events, along with distances between contaminant contributors and receiving waters appeared to influence effectiveness of the holding pond for mitigating downstream bacterial contamination. During 2006 and 2007, the timing of sample collections and nearby presence of cattle appeared to be a key factor influencing *E. coli* counts. These results provided a better understanding of limitations that could affect benefits of a holding pond mitigating bacteriological issues at other locations. Unless additional livestock management considerations are factored into the process, benefits for bacterial mitigation by use of a holding pond may be negated.

4.3.2 Influences on Holding Pond *E. coli* Counts and Die-off

Data from 2006 and 2007, although relatively limited, provided insight on how *E. coli* counts responded within the holding pond and how the magnitude and timing between runoff events could affect functioning of the holding pond. During 2006 relatively high amounts of runoff over a seven day period (between March 31 and April 6) quickly filled the holding pond and caused premature discharge out the emergency overflow spillway. Quick filling of the holding pond appeared to evenly distribute *E. coli* fairly well because counts in the spillway discharge (MS5B) were similar to those in runoff entering the pond at MS5. While the initial *E. coli* sample from the spillway discharge (800 *E. coli* 100 mL⁻¹ at MS5B) collected on April 6 was over two orders of magnitude lower than in runoff entering the pond at MS5 (110,000 *E. coli* 100 mL⁻¹), subsequent samples showed *E. coli* counts between these sites were very similar until approximately April 19 (Figure 7a). The initial lower value from the spillway overflow on April 6 compared to the count at the pond inlet was considered to be due to diluting effects from uncontaminated snow melt-water at the spillway outlet. This snow melt-water mixed with overflow discharge during initial sampling and was mainly gone by the time the next sample was collected from the spillway four days later on April 10, 2006.

Once the holding pond was full, subsequent individual runoff events appeared to have less influence on counts within the holding pond due to the much lower volume of runoff compared to the holding pond volume. This was most evident from April 26 to May 27, 2006. During this period *E. coli* counts remained relatively stable in spillway runoff despite counts in runoff entering the pond being an order of magnitude lower and almost two orders of magnitude higher on April 26 and in late May, respectively (Figure 7a). The low *E. coli* count in runoff from the farmyard-cattle-corral on April 26 was thought to be due to very low flows that caused reduced flushing of *E. coli* from the farmyard site. The hydrograph for MS5 during 2006 showed measured flow diminished substantially after April 12 with rates generally less than one or two litres per second (Appendix 6c). During 2007 runoff also filled the holding pond fairly quickly and within-pond counts (site MS5A) were fairly similar to those in runoff at the inlet (site MS5) by the time the initial runoff period ended around April 3, 2007 (Figure 7b). Overflow out of the emergency spillway had not occurred in 2007 because runoff volumes were less than during 2006 (Appendix 6c). Similar to 2006, *E. coli* counts in runoff entering the pond during 2007 appeared to have minimal influence on within-pond counts once the holding pond had been filled. An elevated *E. coli* count (approximately 46,000 *E. coli* 100 mL⁻¹) in runoff from the farmyard-cattle-corral at site MS5 on May 6, 2007 was several orders of magnitude higher than within-pond counts, yet subsequent samples from within the holding pond at sampling site MS5A over the next couple of days showed minimal effects. Within-pond counts at this time only ranged between 3 *E. coli* 100 mL⁻¹ to 43 *E. coli* 100 mL⁻¹ (Figure 7b). Even though emptying of the holding pond had started much earlier in 2007 (on April 18), within-pond volumes still appeared to be large enough to overcome the influence of the high *E. coli* count in runoff entering the pond. This was probably due to a very low amount of runoff volume associated with this high *E. coli* count (Appendix 6c).

Unlike 2006, there were also more frequent late spring and summer precipitation events in 2007 that further showed how holding pond *E. coli* counts could be greatly influenced by the frequency and size of runoff events. There were four significant precipitation events with peak flows ranging from approximately 15 L s⁻¹ to 40 L s⁻¹ that occurred on May 22 to 24, June 6 to 8, June 23 to 27, and July 10 to 11, 2007 (Appendix 6c). During each event, daily *E. coli* counts from the farmyard-cattle-corral at MS5 (inlet to the holding pond) generally ranged

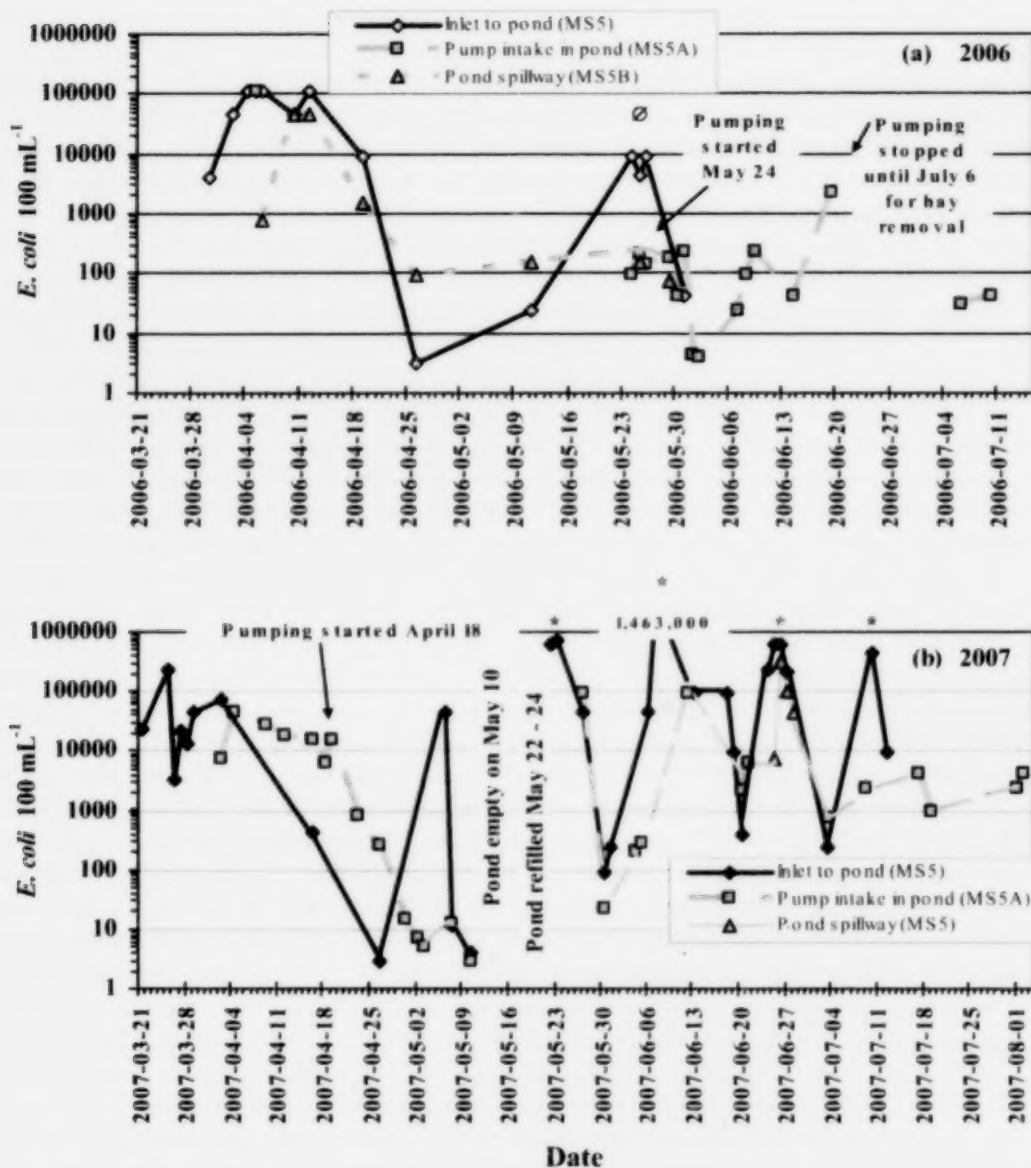


Figure 7. *Escherichia coli* counts ($E. coli$ 100 mL⁻¹) within the holding pond capturing runoff from the farmyard-cattle-corral in the WEBS Stepieler Watershed Project, Manitoba, Canada, 2004 to 2007.

Note: Where more than one *E. coli* sample was collected on a day, points on the graph represent the mean daily count;

⊗ = denotes a 2006 peak flow event that occurred after initial pond fill and that was 10 L s⁻¹ or more;

* = denotes a 2007 peak flow event that occurred after initial pond fill and that was 15 L s⁻¹ or more.

between 430,000 *E. coli* 100 mL⁻¹ to over a million *E. coli* 100 mL⁻¹ (Figure 7b). These high *E. coli* counts appeared to cause similar corresponding increases in holding pond counts, albeit within-pond counts tended to be considerably lower than in runoff at MS5 (Figure 7b). Compared to daily runoff events in early spring 2007 that occurred over approximately two weeks, these multiple episodic runoff events over an approximate 50 day period appeared to slow down die-off within the holding pond by providing interspersed new introductions of fresh *E. coli* every couple of weeks. Similar to previous observations however, as the volume in the holding pond increased, *E. coli* counts in runoff from the farmyard-cattle-corral appeared to have less influence on increasing within-pond counts. This was most evident by the July 10 runoff event where the *E. coli* count in runoff from the farmyard-cattle-corral was 430,000 *E. coli* 100 mL⁻¹ yet within-pond counts only showed a relative marginal increase from approximately 2,300 *E. coli* 100 mL⁻¹ to 4,300 *E. coli* 100 mL⁻¹ (Figure 7b).

The influence of reduced flow on flushing bacteria from the farmyard-cattle-corral location was also evident for each of these runoff events in 2007. *Escherichia coli* counts in farmyard-cattle-corral runoff declined to their lowest observed values on April 26, May 10, May 30, June 20, and July 3 (Figure 7b). These counts coincided with runoff flows diminishing to less than 2 L s⁻¹ (Appendix 6c). The three lowest *E. coli* counts (ranging from 3 to 43 *E. coli* 100 mL⁻¹) in runoff from the farmyard-cattle-corral during 2006 also occurred when flows were 2 L s⁻¹ or less (Figure 7a, Appendix 6c). However, based upon this limited data it is difficult to fully understand what may be a critical flow rate in the Stepper Watershed to cause significant increases in *E. coli* counts from the farmyard-cattle-corral. For example, a modest increase in flow to slightly more than 3 L s⁻¹ on May 6, 2007 caused a significantly elevated *E. coli* count of approximately 46,000 *E. coli* 100 mL⁻¹ in runoff from the farmyard-cattle-corral. This indicated that other factors were important to determining *E. coli* contributions from the farmyard-cattle-corral such as the amount of fresh manure exposed to runoff along with timing between runoff events.

From a holding pond operational perspective, two scenarios of interest included the number of days it took: i) from when spring runoff effectively stopped filling the pond and *E. coli* counts started a steady decline to finally becoming less than 200 *E. coli* 100 mL⁻¹, and ii) from when spring runoff started filling the holding pond to when *E. coli* counts were less than 200 *E. coli* 100 mL⁻¹. Estimates indicated it took about 14 and 24 days from the time spring runoff effectively stopped filling the holding pond to when *E. coli* counts decreased to approximately 200 *E. coli* 100 mL⁻¹ during 2006 and 2007, respectively (Table 4). These estimates were derived from regression analyses on declining *E. coli* counts after the initial spring runoff period basically ended (Appendix 8). In 2006 and 2007 the end of the majority of spring runoff appeared to end around April 12 and April 3, respectively (Figures 7a and 7b). At this time runoff flows had declined to low values that were usually less than a couple of litres per second (Appendix 6c). In 2006, the holding pond started filling with spring runoff on March 30. This was 13 days earlier than when runoff was considered to have stopped filling the pond on April 12. This indicated it took approximately 27 days from when the pond began to fill to when *E. coli* counts were less than 200 *E. coli* 100 mL⁻¹. In 2007, it was estimated to take approximately 37 days from when runoff began to fill the pond on March 21 to when *E. coli* counts declined to 200 *E. coli* 100 mL⁻¹ (Table 4).

While it took a greater number of days for *E. coli* counts to diminish to 200 *E. coli* 100 L⁻¹ in 2007, the count at the end of the initial runoff period had declined to a much lower value than in 2006 (Figures 7a and 7b). This lower *E. coli* count in 2007 compared to 2006 probably occurred because emptying of the holding pond by irrigation pumping had started

Table 4. Number of days for *E. coli* counts within the holding pond to decline to approximately 200 *E. coli* 100 mL⁻¹ or less during spring runoff in the WEBs Stepler Watershed Project, 2004 to 2007.

Year	Scenario	Timeframe	No. of days
2006	Spring runoff effectively stops filling the holding pond to when <i>E. coli</i> counts were approximately 200 <i>E. coli</i> 100 mL ⁻¹ or less	April 12 – April 25	14
	Spring runoff starts filling the holding pond to when <i>E. coli</i> counts were approximately 200 <i>E. coli</i> 100 mL ⁻¹ or less	March 30 – April 25	27
2007	Spring runoff effectively stops filling the holding pond to when <i>E. coli</i> counts were approximately 200 <i>E. coli</i> 100 mL ⁻¹ or less	April 3 – April 26	24
	Spring runoff starts filling the holding pond to when <i>E. coli</i> counts were approximately 200 <i>E. coli</i> 100 mL ⁻¹ or less	March 21 – April 26	37

much earlier in 2007. Initial pumping from the holding pond in 2007 began on April 18 and occurred on a fairly regular basis until the pond was basically empty by May 10. Pumping in 2006 had not started until May 24 so water depth remained higher for a longer interval and may have helped *E. coli* at depth to survive longer. The holding pond had high water colour and turbidity due to the originating runoff mainly coming from the farmyard-cattle-corral. Turbidity in the water was probably enough to reduce the effects of sunlight exposure nearer bottom even though the pond was not much more than a metre deep. Secchi depth measurements taken in May 2007 only ranged from 12 to 13 cm indicating fairly limited light penetration (Appendix 9). The earlier emptying of the pond in 2007 could have increased and quickened exposure of *E. coli* nearer the pond bottom to the sunlight's ultraviolet radiation.

4.3.3 Holding Pond *E. coli* Counts In Irrigation Discharge

Initial emptying of the holding pond via irrigation in 2006 began on May 23. *Escherichia coli* samples were collected in the holding pond near the pump intake and in the field from irrigation ejector nozzles and collection pans set near ejector nozzles to determine if the irrigation pumping process had any measurable effect on *E. coli* counts. Biederbeck (1979) found irrigation pumping of sewage lagoon effluent with a centrifugal pump impeller pressure of approximately 60 pounds per square inch (psi) reduced fecal coliform and fecal streptococci by 66 % and 91 %, respectively. Additional results from laboratory experimentation in this study using a reduced centrifugal pumping pressure of 50 psi showed a reduction in fecal coliform counts of only about 15 % after one pass through the pump. However, there was a cumulative effect of approximately 45 % and 90% on second and fourth passes, respectively.

There was no apparent consistent effect between *E. coli* counts from within the Steppler holding pond to those measured at the irrigation ejectors or field pans during 2006. Daily *E. coli* counts were relatively similar between the three sampling sites and there was no significant difference ($p > 0.23$) between the mean *E. coli* values (Figure 8a). Although daily counts of *E. coli* for the irrigation pan in the field had slightly higher counts than from the holding pond source water on June 12 and July 6, 2006, this was thought to probably be due to the natural variability that often occurs with bacteria enumeration. Other possibilities could be contamination from wildlife such as small birds drinking or bathing in the pans but there were no observations or evidence of that occurring.

Sampling from within the holding pond near the pump and from irrigation ejector nozzles in 2007 showed similar results to 2006 (Figure 8b). *Escherichia coli* counts from these two locations were very similar and not significant ($p > 0.15$) indicating the effects from irrigation pumping and ejection into open air had minimal effect on *E. coli* counts. Irrigation pumping pressure used in the Steppler Watershed was estimated to be about 90 psi at the pump and approximately 55 to 60 psi at the last nozzle. However, this small pump did not appear to cause large enough differential changes in compression and decompression of bacterial cells in the pumping process. Fraser (1951) found as much as 90 % of bacterial cells ruptured from a pressure shock when bacterial cells were compressed and then pressure was suddenly released during decompression. Biederbeck (1979) could not pinpoint causes of fecal coliform destruction during irrigation pumping and thought it was probably due to a strong pressure shock, possible oxygenation and bubble formation, as well as sheering action and cavitation occurring within the centrifugal force field of the pump impeller.

4.3.4 Test-wells for Farmyard-Cattle-Corral Drainage Channel

Testing for *E. coli* in four relatively shallow test-wells (depth of approximately 0.5 m below elevation of water course channel bottom) located near the farmyard-cattle-corral and adjacent to the drainage channel to site MS5 (Figure 4) was conducted to help determine if flow to the holding pond was originating from more than just surface runoff. The holding pond generally filled quicker than predicted for the size of the drainage area and it became apparent that a portion of the flow entering the holding pond was coming from either shallow sub-surface flow or groundwater base flow. Runoff coefficients (runoff volume at MS5/precipitation volume on associated watershed area) from the cattle confined area were often close to 50 % and as high as 66 % during May 22 to 23, 2007 (J. Yarotski pers comm.). Meanwhile, runoff coefficients at other sites within the watershed were generally half this amount due to loss from ground infiltration, evaporation, transpiration, among other factors. Water absence and presence in the test-wells helped determine when sub-surface flow occurred while sampling for *E. coli* presence provided a means of verifying whether surface runoff and associated contaminants from the farmyard-cattle-corral were recently entering into groundwater and being transported through subsurface flow.

Escherichia coli results collected during this study verified that bacterial contamination was occurring in the shallow groundwater flow. Sampling for *E. coli* from the test-wells was first done in mid June 2006. Only 10 samples in total were collected from the four test-wells during 2006. The highest contamination level in 2006 was found in drain test-well DTW2 (Figure 9a). This maximum *E. coli* count occurred on June 19 and was 93 *E. coli* 100 mL⁻¹.

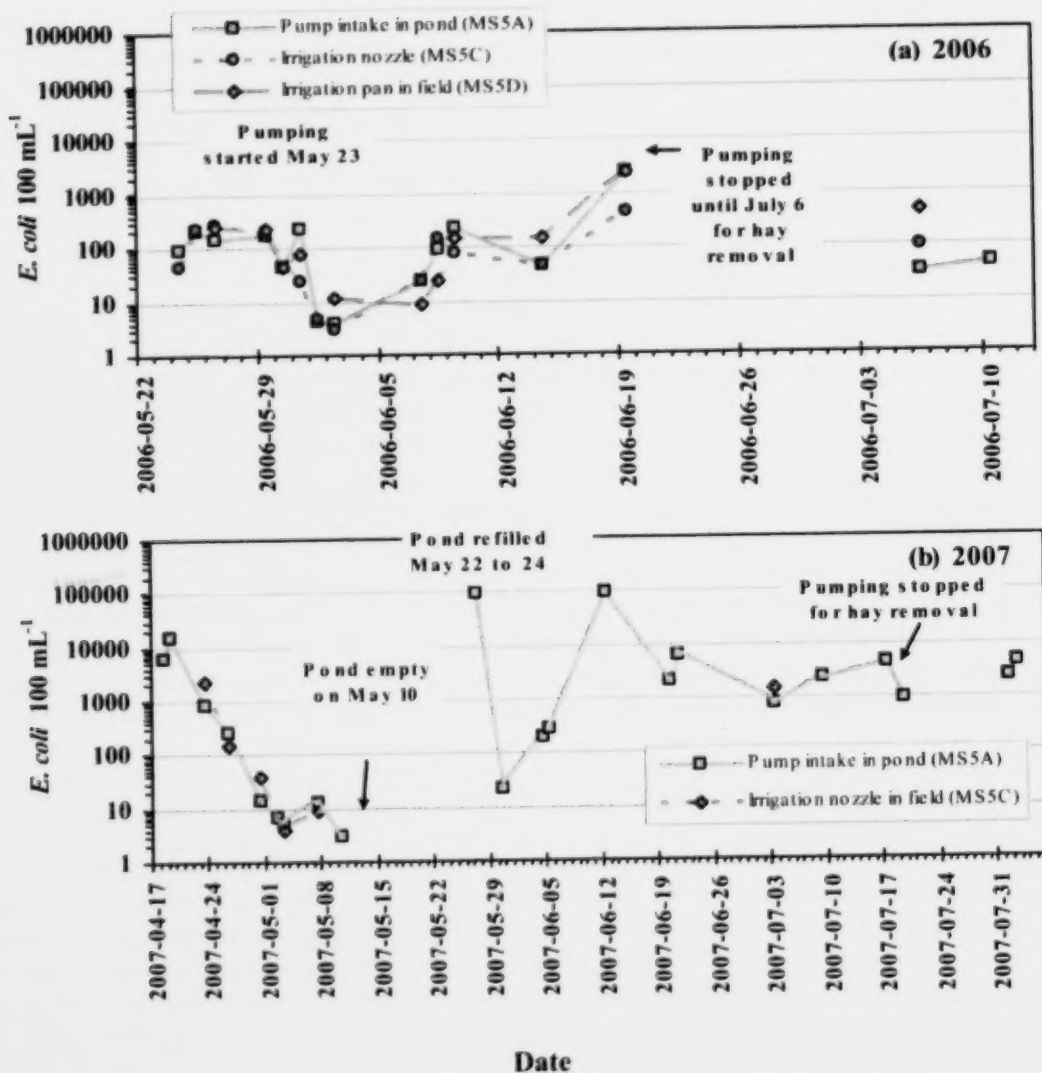


Figure 8. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) within the holding pond and at the ejector nozzles in the field during pond drawdown by irrigation pumping in the WEBs Stepipler Watershed Project, Manitoba, Canada, 2004 to 2007.

There were only two samples collected from each test-well DTW3 and DTW4 in 2006. All counts from these two test-wells were less than the laboratory detection limits.

Sampling from the test-wells was continued in 2007 to validate results from the previous year. However, sampling from DTW3 and DTW4 in 2007 was started in early April with periodic collections up to August 1. Sampling from test-wells DTW1 and DTW2 was less frequent because they often had very little or no water when sampling was occurring at the other test-wells. Field observations at the test-wells indicated that they tended to be dry

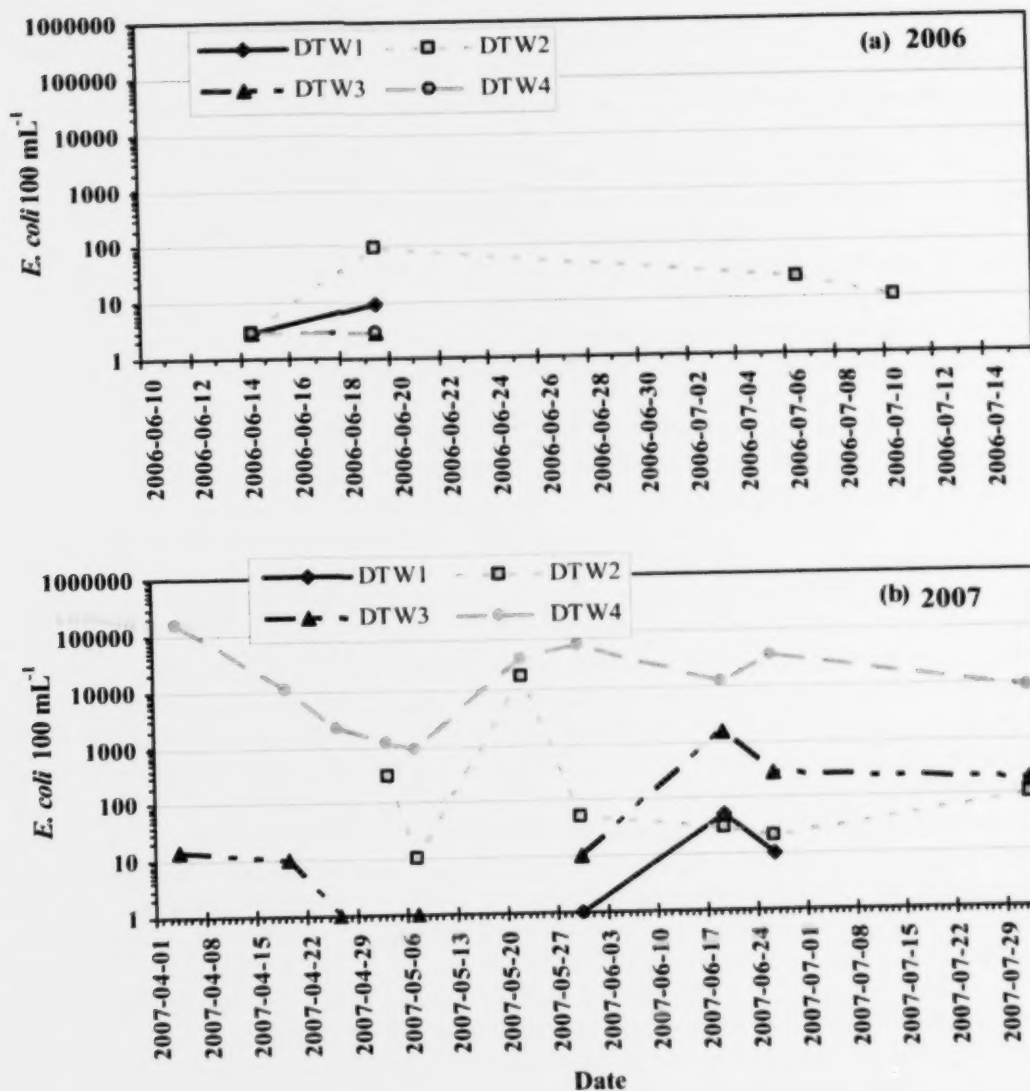


Figure 9. *Escherichia coli* counts ($E. coli$ 100 mL⁻¹) in drain shallow test-well samples from the WEBs Steppler Watershed Project, Manitoba, Canada, 2004 to 2007.

during extended periods without rainfall but they were fairly responsive to precipitation events (B. Turner pers comm.). This indicated subsurface flow was a factor at this location. Unlike 2006, the highest *E. coli* counts during 2007 consistently came from DTW4 (Figure 9b). Counts of *E. coli* from DTW4 ranged from a maximum of 150,000 *E. coli* 100 mL⁻¹ on the first day of sampling (April 4) to a minimum of 880 *E. coli* 100 mL⁻¹ on May 7, 2007. Test-well DTW4 counts in 2007 were significantly different ($p < 0.002$) than samples from the other test-wells. It was also noted in observations recorded during sample collections that water in DTW4 was frequently frothy which further indicated surface water and associated constituents such as nutrients and organic matter was recently reaching this shallow

groundwater zone. *Escherichia coli* counts at DTW4 throughout the rest of sample collections to August 1 continued to be elevated with values ranging from 8,000 *E. coli* 100 mL⁻¹ to 57,000 *E. coli* 100 mL⁻¹ (Figure 9b).

Escherichia coli counts from the other test-wells in 2007 were much lower than observed in DTW4 and generally under 200 *E. coli* 100 mL⁻¹ (the water quality guideline for primary recreational activities). However, even though there were relatively low *E. coli* counts found in these other test-wells, the mere presence of *E. coli* in groundwater indicated contamination from surface sources. Crane and Moore (1984) noted that contamination of groundwater was generally not thought to be a problem because of the soils ability to absorb and filter bacteria from percolating effluents, but they also found bacteria movement was largely based upon soil structure and biological factors such as soil pH, temperature, moisture, and nutrient supply. From their review of research, Hagedorn and McCoy (1979) concluded that bacteria generally moved less than 1 m in unsaturated conditions but could move as far as 30 to 60 m under saturated situations. Various other studies found removal of bacteria to non detectable values from applied effluents ranged from 5 cm to 48 cm below the soil surface (Gerba *et al.* 1975, Dazzo *et al.* 1973), while Stoddard *et al.* (1998) reported fecal coliforms reached 90 cm depths on shallow karst soils applied with dairy manure. Other authors found soil characteristics and structure were a predominant influence on bacterial movement or reductions. Clay content increased bacterial loss over sandy soils while well structured soils containing macropores were less effective bacterial filters than disturbed soils (McGechan and Vinten 2003, Fenlon *et al.* 2000, Smith *et al.* 1985). Finding shallow subsurface flow in the Steppler Watershed shallow test-wells was not unexpected since soils along the Manitoba Escarpment are comprised of till/shale mixes. It was also understandable to find *E. coli* presence in these test-wells since they were only about half a metre to a metre below the soil surface. Water and bacteria in the Steppler Watershed test-wells indicated some surface runoff from the farmyard-cattle-corral was lost to groundwater infiltration, but a good portion was still making its way to the holding pond through shallow subsurface flow and causing more volume than originally expected. This would reduce capacity in the holding pond and probably helped contribute to overflow out of the pond spillway in 2006 that shortened holding times for *E. coli* count reductions.

Contributions to the Steppler Watershed holding pond from deeper groundwater base flow were considered to be minimal. A review of the 2007 hydrograph for site MS5 indicated that base flow probably provides a relatively small proportion compared to surface runoff and shallow sub-surface flow (J. Yarotski pers comm.). Flow peaks in the hydrograph for MS5 coincided with rainfall events and greater amounts of rainfall coincided with more significant flow peaks. The flow to MS5 also responded fairly quickly to near zero once a rainfall event finished (Appendix 6c). If base flow was more predominant, it would be expected that the hydrograph flow would not respond so quickly and continue for an extended period. As well, two *E. coli* samples from deep borehole test-wells near the holding pond (Figure 4) indicated that enteric bacterial contamination was not penetrating into groundwater at depths of approximately 3.9 metres. Sampling from the borehole test-wells BH9 and BH11 had *E. coli* values of < 1 *E. coli* 100 mL⁻¹ (Appendix 4).

4.4 Cattle Winter Feeding Field Site

Runoff samples were collected in the drainage ditch adjacent to a winter feeding field for livestock to determine if significant *E. coli* contamination was occurring to runoff coming in contact with residual manure (cowpies) deposited on the field over winter and early spring. The winter feeding field was adjacent to the Steppler Watershed field F7 but outside of the Steppler Watershed (Figure 2). This field was also where irrigation water from the holding pond was applied when it was emptied in late spring and summer.

Only a very limited number of samples were first collected from an upstream and downstream site in receiving drainage during spring runoff 2006. Results of these samples showed relatively high to very high *E. coli* contributions occurred at the downstream sampling site MS17 during the early stages of spring runoff. On March 30 and April 2, 2006, *E. coli* counts from MS17 were 1,500 *E. coli* 100 mL⁻¹ and > 110,000 *E. coli* 100 mL⁻¹, respectively (Figure 10a). Meanwhile, *E. coli* samples from the upstream site (MS16; Figure 2) were considerably lower and ranged between 220 to 10 *E. coli* 100 mL⁻¹. Subsequent sampling from April 4 to April 6, 2006 indicated that upstream and downstream *E. coli* counts were fairly low with all counts being less than 100 *E. coli* 100 mL⁻¹ (Figure 10a).

While *E. coli* counts observed from the winter feeding area in 2006 appeared to show manure deposited on the winter feeding field could contribute fairly significant amounts of *E. coli* in runoff, it is not known from these few samples how significant the contributions might have been. Although sampling frequency and length of the sampling period increased during 2007, the extent of contributions from the winter feeding site were still difficult to discern. The first sets of samples collected from March 13 to 25, 2007 showed that *E. coli* counts at the upstream site MS16 were often higher than at MS17 (downstream site) but were still generally less than 200 *E. coli* 100 mL⁻¹ (Figure 10b). On March 22, 2007, there was an apparent significant contribution in runoff from the winter feeding site since the bacteria count at MS17 was 430,000 *E. coli* 100 mL⁻¹. Meanwhile, counts were under a 1000 *E. coli* 100 mL⁻¹ at MS16. However, by April 6, 2007, *E. coli* counts at the upstream site MS16 increased significantly to 387,000 *E. coli* 100 mL⁻¹ compared to only 32,000 *E. coli* 100 mL⁻¹ at MS17 (Figure 10b). Counts at MS16 remained elevated above counts at MS17 for the duration of the sampling in 2007.

High *E. coli* counts at MS16 in 2007 were initially unexpected because there had not been any known previous sources that would provide this level of contamination. However, field observations recorded during sample collections in 2007 noted sheep were in a field adjacent and just upstream of MS16 when the sample was collected on June 6. Sheep had been added to the Steppler Watershed farm operation in summer 2006. Similar to what was previously noted in Section 4.3 regarding the influence of nearby cattle presence to *E. coli* counts at sampling sites MS3 and MS4, the proximity of sheep also appeared to have a significant affect on levels of *E. coli* counts transported to receiving waters at sampling site MS16. Elevated counts in runoff from the cattle over-wintering field, especially in 2006 were not surprising given some of the manure on the field was relatively recent and would not have been subjected to prolonged freezing as would have occurred for earlier deposits. Soupir *et al.* (2005) found runoff from a pasture with cowpie treatments had fecal coliform counts of approximately 137,000 organisms 100 mL⁻¹ while fresh cattle manure was found to be a long-term source of bacterial contamination (Thurston-Enriquez *et al.* 2005). While fecal coliforms declined to low levels within 40 d after manure was applied in frozen conditions, Culley and Phillips (1982) also demonstrated longevity of fecal streptococci bacteria in frozen manure.

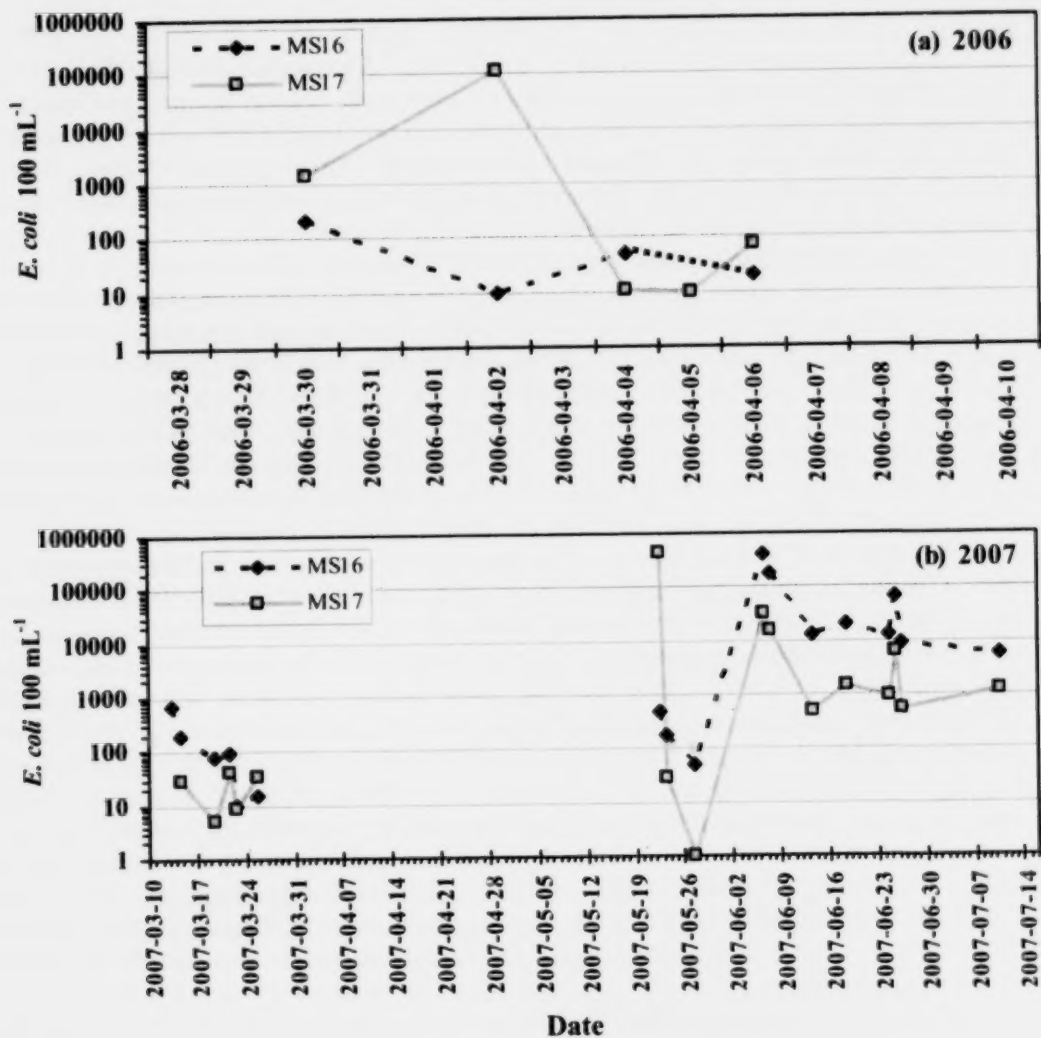


Figure 10. *Escherichia coli* counts ($E. coli$ 100 mL⁻¹) from sampling sites upstream (MS16) and downstream (MS17) of the cattle winter feeding site at the WEBs Steppler Watershed Project, Manitoba, Canada, 2004 to 2007.

Results in 2007 also indicated there was a fairly substantial natural attenuation within the approximate 1000 m between sites MS16 and MS17. Daily *E. coli* counts between June 6 and July 10 at site MS17 were usually an order of magnitude lower than corresponding daily counts upstream at MS16 (Figure 10b, Appendix 4). This indicated that fresh manure deposits from the sheep had a much greater impact to runoff than older manure deposits from the cattle winter feeding site. Fresh cattle feces a couple of days old were also observed to produce significantly higher counts than 100 d old ones in a study conducted in Utah (Kress and Gifford 1984).

4.5 Holding Pond Considerations

Although effectiveness of the holding pond at significantly reducing transport of *E. coli* counts to downstream areas any more than natural attenuation was inconclusive in this study, it did show counts entering the pond during runoff could be substantially reduced prior to release of water from the pond. Counts of *E. coli* entering the holding pond were often in the range of 10^4 to 20^5 *E. coli* 100 mL⁻¹ while counts were orders of magnitude lower as the pond was emptied. A variety of factors such as livestock presence at sites, frequency and magnitude of runoff events, distances between contaminant contributors and receiving waters, among others can have significant influences on the effectiveness of the holding pond for mitigating bacterial contamination.

It has been well documented by numerous authors of the likelihood for elevated levels of microorganisms that includes bacteria, viruses, and other potentially pathogenic organisms to occur in livestock manures. Occasional outbreaks of human illness or fatalities due to links with food or water contaminated from manure sources reinforces the need to avoid too much complacency on the potential human health impacts that can arise from this source. While reported human infections usually occur due to incidences such as consuming inadequately washed food fertilized with cattle manure (Chapman *et al.* 1997, Cieslak *et al.* 1993), the Walkerton, Ontario incident in 2000 also marked how external bacterial inputs to water supplies and a corresponding breakdown in the municipal drinking water treatment process can also be devastating (O'Connor 2002). These types of incidences are reminders on the importance to develop and implement safe, efficient manure management techniques.

Use of a holding pond to capture contaminated runoff and provide some mitigating treatment prior to release to downstream waters is one mechanism that may help reduce potential impacts from bacterial and other pathogenic contamination. However, based upon results from the 2004 to 2006 Stepler Watershed data, benefits of establishing a holding pond to mitigate bacterial contamination should consider site specific conditions. For *E. coli* contamination issues, a holding pond may have greater benefits in situations where contaminated runoff has limited exposure to the sunlight's ultraviolet radiation and much shorter travel times to receiving lakes, streams, and rivers than in the Stepler Watershed. There appeared to be reasonably good natural attenuation of *E. coli* in this watershed due to the intermittent nature of water courses and the relatively long distances before contaminated runoff entered permanent downstream waters. It must also be remembered though, that *E. coli* is a surrogate for the potential presence of other more harmful pathogens. Attenuation of other pathogens in some cases may not necessarily be as great as observed for *E. coli* so it would be still considered prudent to minimize contamination potential as much as possible.

5.0 CONCLUSIONS

- 1) Mean *E. coli* counts from most sites within the Steppler Watershed were usually less than the water quality guideline limit for primary recreational activity that is a maximum of 200 *E. coli* 100 mL⁻¹;
- 2) Mean *E. coli* counts in runoff from the farmyard-cattle-corral were significantly higher than the primary recreational guideline value and usually significantly higher than at other sampling locations;
- 3) The maximum *E. coli* count each year also came from the farmyard-cattle-corral and was as high as 2,300,000 *E. coli* 100 mL⁻¹;
- 4) The minimum mean *E. coli* count was observed at the background site located in a natural wooded watershed with maximum counts from this watershed being less than 100 *E. coli* 100 mL⁻¹;
- 5) Occasional very high *E. coli* counts occurred in the Steppler Watershed at some sampling sites other than from the farmyard-cattle-corral and these incidences generally appeared to directly coincide with the presence of livestock nearby these sites during runoff events;
- 6) Incidences of high *E. coli* counts at different sites linked to nearby presence of cattle or sheep during runoff events indicated how important timing and location for livestock management would be for some BMPs to be effective;
- 7) The holding pond was generally effective at intercepting and preventing runoff that was highly contaminated with *E. coli* from being transported to downstream areas. Counts of *E. coli* entering the holding pond were often in the range of 10⁴ to 20⁵ *E. coli* 100 mL⁻¹ while counts were one or more orders of magnitude lower when the pond was being emptied;
- 8) Effectiveness of the holding pond to cause significant decreases in *E. coli* counts any more than natural attenuation in the Steppler Watershed was inconclusive from the 2004 to 2007 data. Mean *E. coli* counts between sites upstream and downstream of the farmyard-cattle-corral were not significantly different prior to or after the holding pond began operating;
- 9) Substantial natural attenuation of *E. coli* counts appeared to occur between the farmyard-cattle-corral site and the receiving downstream site that may have been due to the fairly long distance of approximately 500 m between these locations. This distance provided opportunity for additional dilution and increased potential die-off of *E. coli* from exposure to the sunlight's ultraviolet radiation;
- 10) Significant increases in *E. coli* counts in runoff from the farmyard-cattle-corral generally coincided with more major runoff events that were greater than 2 L s⁻¹;
- 11) As the holding pond volume increased, the relatively much smaller volume from late spring runoff events from the farmyard-cattle-corral appeared to have less influence on within-pond *E. coli* counts than earlier in the spring runoff season;
- 12) It took approximately 15 to 24 days from the time spring runoff effectively stopped filling the holding pond to when *E. coli* counts within the pond were less than 200 *E. coli* 100 mL⁻¹ during 2006 and 2007, respectively;
- 13) It took approximately 28 to 39 days from the time spring runoff started filling the holding pond to when *E. coli* counts within the pond were less than 200 *E. coli* 100 mL⁻¹ during 2006 and 2007, respectively;

- 14) There was no apparent effect or reduction to *E. coli* counts when passed through a small irrigation pump to empty the holding pond.

6.0 RECOMMENDATIONS

- 1) To better understand the effectiveness of the holding pond on *E. coli* count reductions to downstream areas, cattle or other livestock would need to be kept away from upstream and downstream sampling locations during the spring runoff period;
- 2) Upstream and downstream sampling sites closer to the confluence with the channel discharging from site MS5 (holding pond) would probably be needed for better evaluation of the holding pond effectiveness;
- 3) A greater number of sample collections should be considered during a spring runoff season to increase analytical and statistical robustness;
- 4) Sampling for *E. coli* at sites upstream (MS16) and downstream (MS17) of the cattle wintering feeding area should be discontinued while livestock such as sheep are present in fields adjacent to MS16 during the spring runoff period;

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9.0 Appendices

Appendix 1. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) collected from the Webs Steppeler Watershed Project, Manitoba, Canada, 2004.

Sample site reference	Sample site	date (y/m/d)	time (h)	<i>E. coli</i>	
				(<i>E. coli</i> 100 mL ⁻¹)	(log ₁₀)
Background site (Natural Wooded Area)	MS15	2004-04-04	13:20	< 3	0.4771
		2004-04-14	12:00	< 3	0.4771
		2004-04-14	12:00	< 3	0.4771
		2004-05-31	10:48	93	1.9685
		2004-08-10	12:45	99	1.9956
		2004-09-16	17:00	23	1.3617
		2004-09-21	16:00	23	1.3617
		2004-10-20	15:05	24	1.3802
Background site (Madill Watershed)	MS13	2004-03-25	16:55	4,300	3.6335
		2004-03-29	15:00	150	2.1761
		2004-03-31	16:00	< 3	0.4771
		2004-04-04	14:15	15	1.1761
		2004-04-06	12:00	< 3	0.4771
		2004-04-14	13:05	4	0.6021
		2004-04-26	10:45	4	0.6021
		2004-05-16	10:30	43	1.6335
		2004-05-17	10:00	9	0.9542
		2004-05-31	10:00	75	1.8751
		2004-06-07	11:40	930	2.9685
		2004-08-10	13:45	192	2.2833
		2004-09-16	16:30	230	2.3617
		2004-09-21	15:45	230	2.3617
		2004-10-20	14:45	40	1.6021
Background site (Madill Watershed)	MS14	2004-03-28	13:55	230	2.3617
		2004-03-29	16:00	230	2.3617
		2004-03-31	15:35	4	0.6021
		2004-04-04	14:30	< 3	0.4771
		2004-04-06	11:45	4	0.6021
		2004-04-14	13:25	< 3	0.4771
		2004-04-26	11:00	< 3	0.4771
		2004-05-16	10:15	380	2.5798
		2004-05-17	9:45	150	2.1761
		2004-05-31	9:30	230	2.3617
		2004-06-07	12:00	2,300	3.3617
		2004-08-10	13:45	303	2.4814
		2004-09-16	16:30	4	0.6021
		2004-09-21	15:45	230	2.3617

Appendix 1 continued. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) collected from the WEBs Stepler Watershed Project, Manitoba, Canada, 2004.

Sample site reference	Sample site	date (y/m/d)	time (h)	<i>E. coli</i>	
				(<i>E. coli</i> 100 mL ⁻¹)	(log ₁₀)
Upstream site for field F3	MS1	2004-03-28	13:55	230	2.3617
		2004-03-29	12:50	1,200	3.0792
		2004-03-30	9:55	100	2.0000
		2004-03-31	15:05	43	1.6335
		2004-04-06	12:45	4	0.6021
		2004-05-16	10:45	15	1.1761
		2004-05-17	10:15	4	0.6021
		2004-05-31	11:15	43	1.6335
		2004-06-07	12:10	23	1.3617
Downstream site for field F3	MS3	2004-03-28	12:55	230	2.3617
		2004-03-29	12:50	43	1.6335
		2004-03-30	10:50	70	1.8451
		2004-03-31	15:00	38	1.5798
		2004-04-01	17:25	93	1.9685
		2004-04-04	11:30	4	0.6021
		2004-04-06	12:45	23	1.3617
		2004-04-14	14:20	< 3	0.4771
		2004-04-26	11:30	< 3	0.4771
		2004-05-16	11:35	< 3	0.4771
		2004-05-17	11:20	< 3	0.4771
		2004-05-31	12:00	230	2.3617
		2004-06-07	12:30	230	2.3617
		2004-08-10	12:30	128	2.1072
		2004-09-16	10:30	230	2.3617
		2004-09-21	15:15	23	1.3617
		2004-10-20	10:30	6	0.7782
Upstream site for field F4	MS2	2004-03-28	13:15	430	2.6335
		2004-03-29	13:00	210	2.3222
		2004-03-30	10:03	40	1.6021
		2004-03-31	15:10	43	1.6335
		2004-04-01	17:45	93	1.9685
		2004-04-04	11:45	7	0.8451
		2004-04-06	12:25	< 3	0.4771
		2004-04-14	13:40	< 3	0.4771
		2004-05-16	11:25	< 3	0.4771
		2004-05-17	10:30	< 3	0.4771
		2004-05-31	11:50	43	1.6335
		2004-06-07	12:15	23	1.3617

Appendix 1 continued. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) collected from the WEBs Stepler Watershed Project, Manitoba, Canada, 2004.

Sample site reference	Sample site	date (y/m/d)	time (h)	<i>E. coli</i>	
				(<i>E. coli</i> 100 mL ⁻¹)	(log ₁₀)
Downstream site for field F4	MS4	2004-03-28	12:45	230	2.3617
		2004-03-29	12:45	75	1.8751
		2004-03-30	10:30	160	2.2041
		2004-03-31	15:00	230	2.3617
		2004-04-01	17:30	43	1.6335
		2004-04-04	11:30	< 3	0.4771
		2004-04-06	12:35	< 3	0.4771
		2004-04-14	14:10	93	1.9685
		2004-04-26	11:50	43	1.6335
		2004-05-16	11:25	9	0.9542
		2004-05-17	10:45	43	1.6335
		2004-05-31	12:00	930	2.9685
		2004-06-07	12:30	1,200	3.0792
		2004-08-10	12:30	149	2.1732
Downstream site for farmyard-cattle-corral (holding pond inlet area)	MS5	2004-03-25	17:45	230,000	5.3617
		2004-03-25	18:00	93	1.9685
		2004-03-28	12:00	430,000	5.6335
		2004-03-29	11:55	2,300	3.3617
		2004-03-30	11:25	> 110,000	5.0414
		2004-03-30	16:20	2,400	3.3802
		2004-03-31	15:30	1,500	3.1761
		2004-04-01	17:30	46,000	4.6628
		2004-04-06	13:00	230	2.3617
		2004-05-16	11:45	110,000	5.0414
		2004-05-17	11:00	46,000	4.6628
		2004-05-31	11:55	> 110,000	5.0414
		2004-06-07	13:30	9,300	3.9685
		2004-08-10	14:15	46,000	4.6628
		2004-09-16	9:30	2,300	3.3617
		2004-09-21	15:00	110,000	5.0414
		2004-10-20	12:20	9,300	3.9685

Appendix 1 continued. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) collected from the WEBs Stepler Watershed Project, Manitoba, Canada, 2004.

Sample site reference	Sample site	date (y/m/d)	time (h)	<i>E. coli</i>	
				(<i>E. coli</i> 100 mL ⁻¹)	(log ₁₀)
Downstream site of MS3, MS4, and MS5	MS7	2004-03-25	16:25	230	2.3617
		2004-03-28	11:30	2,300	3.3617
		2004-03-29	11:30	1,500	3.1761
		2004-03-30	11:50	410	2.6128
		2004-03-31	14:40	43	1.6335
		2004-04-01	14:40	75	1.8751
		2004-04-04	11:05	43	1.6335
		2004-04-06	13:15	23	1.3617
		2004-04-14	14:25	< 3	0.4771
		2004-04-26	12:20	< 3	0.4771
		2004-05-16	12:00	15,000	4.1761
		2004-05-17	11:30	930	2.9685
		2004-05-31	8:30	47,000	4.6721
		2004-06-07	12:55	4,300	3.6335
		2004-08-10	12:10	510	2.7076
		2004-09-16	9:00	230	2.3617
		2004-09-21	14:30	43	1.6335
		2004-10-20	13:45	352	2.5465
Downstream site for field F7	MS6	2004-03-25	14:36	2,100	3.3222
		2004-03-28	11:20	4,300	3.6335
		2004-03-29	11:40	3,800	3.5798
		2004-03-30	11:40	1,330	3.1239
		2004-03-31	14:45	43	1.6335
		2004-05-31	8:30	93	1.9685
		2004-06-07	12:50	4,300	3.6335

Appendix 1 continued. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) collected from the WEBs Stepler Watershed Project, Manitoba, Canada, 2004.

Sample site reference	Sample site	date (y/m/d)	time (h)	<i>E. coli</i>	
				(<i>E. coli</i> 100 mL ⁻¹)	(log ₁₀)
Downstream site from MS7 and MS6	MS9	2004-03-28	11:12	2,300	3.3617
		2004-03-29	14:00	750	2.8751
		2004-03-30	12:15	500	2.6990
		2004-03-31	14:30	93	1.9685
		2004-04-01	14:25	93	1.9685
		2004-04-04	10:45	210	2.3222
		2004-04-06	13:55	7	0.8451
		2004-04-14	14:55	4	0.6021
		2004-04-26	13:00	< 3	0.4771
		2004-05-16	12:30	9,300	3.9685
		2004-05-17	11:55	2,300	3.3617
		2004-05-31	9:00	7,500	3.8751
		2004-06-07	13:05	2,300	3.3617
		2004-09-16	8:45	93	1.9685
		2004-09-21	14:00	2,100	3.3222
		2004-10-20	13:20	132	2.1206
Downstream site for field F9	MS8	2004-03-25	14:22	3	0.4771
		2004-03-28	10:40	150	2.1761
		2004-03-31	14:00	< 3	0.4771
		2004-05-31	9:10	4	0.6021
Stepler Dam outlet	MS12	2004-03-28	10:55	430	2.6335
		2004-03-30	12:00	380	2.5798
		2004-03-31	14:20	93	1.9685
		2004-04-01	14:35	43	1.6335
		2004-04-04	11:00	4	0.6021
		2004-04-06	13:53	< 3	0.4771
		2004-04-14	14:45	9	0.9542
		2004-04-26	12:45	< 3	0.4771
		2004-05-16	12:15	430	2.6335
		2004-05-17	11:40	9,300	3.9685
		2004-05-31	8:40	430	2.6335
		2004-06-07	13:00	< 3	0.4771
		2004-09-21	14:15	93	1.9685

Appendix 2. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) collected from the Webs Steppler Watershed Project, Manitoba, Canada, 2005.

Sample site reference	Sample site	date (y/m/d)	time (h)	<i>E. coli</i>	
				(<i>E. coli</i> 100 mL ⁻¹)	(log ₁₀)
Background site	MS15	2005-04-07	15:45	overgrown	-
		2005-04-14	15:10	< 10	1.0000
		2005-04-18	16:30	< 10	1.0000
		2005-05-09	15:25	< 10	1.0000
		2005-05-30	15:10	5	0.6990
		2005-06-12		20	1.3010
Background site	MS13	2005-04-18	17:00	< 10	1.0000
Background site	MS14	2005-04-18	17:00	10	1.0000
Downstream site for field F3	MS3	2005-11-02	15:20	< 10	1.0000
Downstream site for field F4	MS4	2005-04-03	12:30	overgrown	-
		2005-04-04	13:30	overgrown	-
		2005-04-05	14:08	< 10	1.0000
		2005-04-06	14:05	10	1.0000
		2005-04-07	13:00	3	0.4771
		2005-04-12	15:30	overgrown	-
		2005-04-14		< 10	1.0000
		2005-04-18	16:45	< 10	1.0000
		2005-05-09	14:30	10	1.0000
		2005-05-23	14:45	15	1.1761
		2005-05-23	13:40	overgrown	-
		2005-05-30	14:50	2,300	3.3617
		2005-06-09	11:50	2,300	3.3617
		2005-06-12		750	2.8751
		2005-06-14	13:20	930	2.9685
		2005-07-03	11:55	230	2.3617
		2005-07-11	10:55	140	2.1461

Field notes on livestock presence:

May 26 - cattle moved into riparian area above and below MS3 and MS4

June 8 - cattle moved from area upstream of MS4

Appendix 2 continued. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) collected from the WEBs Stepler Watershed Project, Manitoba, Canada, 2005.

Sample site reference	Sample site	date (y/m/d)	time (h)	<i>E. coli</i>	
				(<i>E. coli</i> 100 mL ⁻¹)	(log ₁₀)
Downstream site for farmyard- cattle-corral (holding pond inlet area)	MS5	2005-03-30	13:45	25	1.3979
		2005-03-31	17:00	24,000	4.3802
		2005-03-31	11:50	46,000	4.6628
		2005-04-03	12:10	46,000	4.6628
		2005-04-04	13:00	24,000	4.3802
		2005-04-05	13:55	110,000	5.0414
		2005-04-06	14:05	110,000	5.0414
		2005-04-07	14:10	> 110,000	5.0414
		2005-04-12	14:40	> 110,000	5.0414
		2005-04-14	13:20	> 110,000	5.0414
		2005-04-18	16:35	24,000	4.3802
		2005-05-09	13:25	24,000	4.3802
		2005-05-23	13:00	overgrown	-
		2005-05-30	14:00	2,300	3.3617
		2005-06-09	12:00	> 110,000	5.0414
		2005-06-12		110,000	5.0414
		2005-06-14	13:35	> 110,000	5.0414
		2005-07-03	12:10	> 110,000	5.0414
		2005-07-03	14:00	430,000	5.6335
		2005-10-12	14:00	3,800	3.5798
		2005-11-02	14:25	43	1.6335
		2005-11-23	10:45	43	1.6335

Appendix 2 continued. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) collected from the WEBs Stepler Watershed Project, Manitoba, Canada, 2005.

Sample site reference	Sample site	date (y/m/d)	time (h)	<i>E. coli</i>	
				(<i>E. coli</i> 100 mL ⁻¹)	(log ₁₀)
Downstream site of MS3, MS4, and MS5	MS7	2005-03-30	17:00	< 1	0.0000
		2005-03-31	10:30	overgrown	-
		2005-03-31	16:30	overgrown	-
		2005-04-03	11:45	overgrown	-
		2005-04-04	13:30	overgrown	-
		2005-04-05	13:35	60	1.7782
		2005-04-06	13:05	180	2.2553
		2005-04-07	14:50	18	1.2553
		2005-04-12	14:15	overgrown	-
		2005-04-14	14:00	10	1.0000
		2005-04-18	16:20	< 10	1.0000
		2005-05-09	14:00	70	1.8451
		2005-05-23	13:15	1,270	3.1038
		2005-05-30	13:50	35	1.5441
		2005-06-09	12:45	780	2.8921
		2005-06-12		1,500	3.1761
		2005-06-14	13:50	9,300	3.9685
		2005-07-03	13:30	40	1.6021
		2005-07-03	13:30	250	2.3979
		2005-10-12	14:15	510	2.7076
Downstream site for field F7	MS6	2005-03-31	11:45	< 1	0.0000
		2005-03-31	16:00	9	0.9542

Field notes on livestock presence:

- June 2 - cattle moved into Field F5 upstream of MS7 (east of road; see Figure 2)
- June 12 - cattle immediately upstream of MS7 (west of road; see Figure 2)
- June 14 - cattle presence less since they are accessing paddock outside of WEBs
- June 17 - cattle immediately upstream of MS7
- July 3 - cattle in paddock upstream of MS7 but at far end of paddock

Appendix 2 continued. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) collected from the WEBs Stepler Watershed Project, Manitoba, Canada, 2005.

Sample site reference	Sample site	date (y/m/d)	time (h)	<i>E. coli</i>	
				(<i>E. coli</i> 100 mL ⁻¹)	(log ₁₀)
Downstream site from MS7 and MS6	MS9	2005-04-03	11:35	overgrown	-
		2005-04-04	13:30	overgrown	-
		2005-04-05	13:25	70	1.8451
		2005-04-06	15:50	50	1.6990
		2005-04-07	14:40	12	1.0792
		2005-04-12	14:00	overgrown	-
		2005-04-14	12:00	10	1.0000
		2005-04-18	13:40	< 10	1.0000
		2005-05-09	13:45	10	1.0000
		2005-05-23	13:00	overgrown	-
		2005-05-30	13:35	6	0.7782
		2005-06-09	13:30	430	2.6335
		2005-06-12		520	2.7160
		2005-06-14	14:00	320	2.5051
		2005-07-03	12:45	60	1.7782
		2005-07-03	12:01	560	2.7482
		2005-10-12	14:30	250	2.3979
		2005-11-02	15:00	< 10	1.0000
Stepler Dam outlet	MS12	2005-04-03	11:10	overgrown	-
		2005-04-04	13:30	overgrown	-
		2005-04-05	13:10	190	2.2788
		2005-04-06	13:15	600	2.7782
		2005-04-07	14:20	17	1.2304
		2005-04-12	13:00	overgrown	-
		2005-04-14	12:40	70	1.8451
		2005-04-18	13:25	< 10	1.0000
		2005-05-09	13:55	490	2.6902
		2005-05-23	13:00	100	2.0000
		2005-05-30	13:45	1	0.0000
		2005-06-09	13:15	110	2.0414
		2005-06-12		150	2.1761
		2005-06-14	14:10	940	2.9731
		2005-07-03	12:55	120	2.0792
		2005-07-03	13:00	330	2.5185
		2005-10-12	14:40	overgrown	-
		2005-11-02	15:10	9	0.9542

Appendix 3. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) collected from the Webs Steppler Watershed Project, Manitoba, Canada, 2006.

Sample site reference	Sample site	date (y/m/d)	time (h)	<i>E. coli</i>	
				(<i>E. coli</i> 100 mL ⁻¹)	(log ₁₀)
Downstream site for field F3	MS3	2006-05-25	9:20	1,500	3.1761
		2006-05-29	9:15	overgrown	-
Downstream site for field F4	MS4	2006-04-05	15:00	10	1.0000
		2006-04-06	14:30	10	1.0000
		2006-04-10	14:20	30	1.4771
		2006-04-12	15:00	20	1.3010
		2006-04-19	14:00	< 10	1.0000
		2006-04-26	13:30	< 10	1.0000
		2006-05-11	16:00	< 10	1.0000
		2006-05-25	9:00	110,000	5.0414
Downstream site for farmyard-cattle-corral (holding pond inlet area)	MS5	2006-05-29	9:00	overgrown	-
		2006-03-30	14:15	4,120	3.6149
		2006-04-02	12:06	46,000	4.6628
		2006-04-04	14:20	> 110,000	5.0414
		2006-04-05	14:20	> 110,000	5.0414
		2006-04-06	14:40	> 110,000	5.0414
		2006-04-10	10:15	46,000	4.6628
		2006-04-12	16:00	110,000	5.0414
		2006-04-19	13:30	9,300	3.9685
		2006-04-26	12:30	3	0.4771
		2006-05-11	14:30	23	1.3617
		2006-05-24	14:04	9,300	3.9685
		2006-05-25	8:00	4,300	3.6335
		2006-05-29	8:30	9,300	3.9685
		2006-05-31	13:00	43	1.6335

Field notes on livestock presence:

- May 19 - cattle in riparian area of fields F3 and F4 upstream of MS3 and MS4
- May 26 - cattle (77 cows plus calves) moved to yard area for sorting
- May 27 - cattle moved to summer pastures outside the Steppler Watershed
- Aug. 1 - cattle moved to riparian areas in fields F3 and F4 upstream of MS3 and MS4
- Aug. 16 - cattle moved out of riparian areas in fields F3 and F4

Appendix 3 continued. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) collected from the WEBs
 Stepler Watershed Project, Manitoba, Canada, 2006.

Sample site reference	Sample site	date (y/m/d)	time (h)	<i>E. coli</i>	
				(<i>E. coli</i> 100 mL ⁻¹)	(log ₁₀)
Holding pond near irrigation pump intake	MS5A	2006-04-05	14:25	> 110,000	5.0414
		2006-04-05	14:30	> 110,000	5.0414
		2006-04-05	14:35	> 110,000	5.0414
		2006-05-24	14:15	93	1.9685
		2006-05-25	12:45	210	2.3222
		2006-05-26	11:00	170	2.2304
		2006-05-26	15:30	120	2.0792
		2006-05-29	8:30	150	2.1761
		2006-05-29	15:30	210	2.3222
		2006-05-30	12:20	43	1.6335
		2006-05-31	13:00	230	2.3617
		2006-06-01	8:50	4	0.6021
		2006-06-01	8:30	7	0.8451
		2006-06-01	13:40	< 3	0.4771
		2006-06-01	13:50	4	0.6021
		2006-06-02	9:15	< 3	0.4771
		2006-06-02	9:25	9	0.9542
		2006-06-02	14:20	< 3	0.4771
		2006-06-02	14:30	< 3	0.4771
		2006-06-07	14:40	23	1.3617
		2006-06-08	14:15	93	1.9685
		2006-06-09	14:15	230	2.3617
		2006-06-14	15:30	43	1.6335
		2006-06-19	12:40	2,300	3.3617
		2006-07-06	13:00	23	1.3617
		2006-07-06	15:20	43	1.6335
		2006-07-10	13:45	43	1.6335
Holding pond spillway overflow	MS5B	2006-04-06	14:40	800	2.9031
		2006-04-10	15:50	46,000	4.6628
		2006-04-12	13:40	46,000	4.6628
		2006-04-19	13:30	1,500	3.1761
		2006-04-26	12:30	93	1.9685
		2006-05-11	14:30	150	2.1761
		2006-05-25	7:45	150	2.1761
		2006-05-29	7:30	75	1.8751

Appendix 3 continued. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) collected from the WEBs
 Steppler Watershed Project, Manitoba, Canada, 2006.

Sample site reference	Sample site	date (y/m/d)	time (h)	<i>E. coli</i>	
				(<i>E. coli</i> 100 mL ⁻¹)	(log ₁₀)
Irrigation sprinkler nozzle discharge	MS5C	2006-05-24	14:30	43	1.6335
		2006-05-25	14:00	230	2.3617
		2006-05-26	12:00	380	2.5798
		2006-05-26	15:35	200	2.3010
		2006-05-29	10:30	93	1.9685
		2006-05-29	15:45	430	2.6335
		2006-05-30	12:35	43	1.6335
		2006-05-31	13:20	23	1.3617
		2006-06-01	9:00	< 3	0.4771
		2006-06-01	14:05	4	0.6021
		2006-06-01	14:15	9	0.9542
		2006-06-02	10:30	3	0.4771
		2006-06-02	14:45	3	0.4771
		2006-06-07	15:00	23	1.3617
		2006-06-08	14:30	150	2.1761
		2006-06-09	14:00	75	1.8751
		2006-06-14	15:40	43	1.6335
		2006-06-19	12:40	430	2.6335
		2006-07-06	15:20	93	1.9685
Irrigation sprinkler test field pan	MS5D	2006-05-26	12:10	280	2.4472
		2006-05-26	15:45	210	2.3222
		2006-05-29	10:40	230	2.3617
		2006-05-29	15:55	230	2.3617
		2006-05-30	12:45	43	1.6335
		2006-05-31	13:30	75	1.8751
		2006-06-02	10:40	9	0.9542
		2006-06-02	14:55	15	1.1761
		2006-06-07	15:10	9	0.9542
		2006-06-08	14:40	23	1.3617
		2006-06-09	14:10	150	2.1761
		2006-06-14	15:50	150	2.1761
		2006-06-19	13:00	2,300	3.3617
		2006-07-06	15:30	430	2.6335

Appendix 3 continued. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) collected from the WEBs
WEBs Stepler Watershed Project, Manitoba, Canada, 2006.

Sample site reference	Sample site	date (y/m/d)	time (h)	<i>E. coli</i>	
				(<i>E. coli</i> 100 mL ⁻¹)	(log ₁₀)
Downstream site of MS3, MS4, and MS5	MS7	2006-04-04	14:20	250	2.3979
		2006-04-05	13:05	1,450	3.1614
		2006-04-06	13:44	80	1.9031
		2006-04-10	13:45	80	1.9031
		2006-04-12	16:00	50	1.6990
		2006-04-19	13:00	< 10	1.0000
		2006-04-26	13:15	< 10	1.0000
		2006-05-11	15:00	< 10	1.0000
		2006-05-25	10:06	2,900	3.4624
		2006-05-29	9:30	50	1.6990
		2006-06-01	13:30	230	2.3617
Downstream site for field F7	MS6	2006-04-05	13:00	30	1.4771
Downstream site from MS7 and MS6	MS9	2006-04-04	14:20	890	2.9494
		2006-04-06	13:00	170	2.2304
		2006-04-10	12:55	70	1.8451
		2006-04-12	13:50	30	1.4771
		2006-04-19	12:50	< 10	1.0000
		2006-04-26	13:00	< 10	1.0000
		2006-05-11	15:15	< 10	1.0000
		2006-05-25	12:15	2,700	3.4314
		2006-05-29	9:45	290	2.4624
		2006-06-01	13:20	50	1.6990
Stepler Dam outlet	MS12	2006-04-04	14:20	400	2.6021
		2006-04-05	13:40	40	1.6021
		2006-04-06	12:40	70	1.8451
		2006-04-10	12:45	260	2.4150
		2006-04-12	13:40	200	2.3010
		2006-04-19	12:55	< 10	1.0000
		2006-04-26	12:45	< 10	1.0000
		2006-05-11	15:30	< 10	1.0000
		2006-05-25	12:05	120	2.0792
		2006-05-29	9:50	20	1.3010
		2006-06-01	13:25	< 3	0.4771

Field notes on livestock presence:

May 19 - cattle in west end of Field F5 upstream of MS7

May 26 - cattle (77 cows plus calves) moved to yard area for sorting

May 29 - 40 cows plus calves moved to east riparian area in Field F5 upstream of MS7

Aug. 1 - cattle moved to Field F5 upstream of MS7 and riparian areas of fields F3 and F4

Aug. 16 - cattle moved out of Field F5 and riparian areas in fields F3 and F4

Appendix 3 continued. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) collected from the WEBs
Steppler Watershed Project, Manitoba, Canada, 2006.

Sample site reference	Sample site	date (y/m/d)	time (h)	<i>E. coli</i>	
				(<i>E. coli</i> 100 mL ⁻¹)	(log ₁₀)
Upstream ditch site from winter holding area	MS16	2006-03-30	14:15	220	2.3424
		2006-04-02	12:06	10	1.0000
		2006-04-04	14:20	50	1.6990
		2006-04-06	14:15	20	1.3010
Downstream ditch site from winter holding area	MS17	2006-03-30	14:15	1,500	3.1761
		2006-04-02	12:06	> 110,000	5.0414
		2006-04-04	14:20	11	1.0414
		2006-04-05	13:50	10	1.0000
		2006-04-06	14:10	70	1.8451
Confined area drainage ditch test-well #1	DTW1	2006-06-14	15:00	< 3	0.4771
		2006-06-19	11:00	9	0.9542
Confined area drainage ditch test-well #2	DTW2	2006-06-14	15:00	< 3	0.4771
		2006-06-19	11:00	93	1.9685
		2006-07-06	12:30	23	1.3617
		2006-07-10	13:45	< 10	1.0000
Confined area drainage ditch test-well #3	DTW3	2006-06-14	15:00	< 3	0.4771
		2006-06-19	11:00	< 3	0.4771
Confined area drainage ditch test-well #4	DTW4	2006-06-14	15:00	< 3	0.4771
		2006-06-19	11:00	< 3	0.4771

Appendix 4. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) collected from the Webs Steppler Watershed Project, Manitoba, Canada, 2007.

Sample site reference	Sample site	date (y/m/d)	time (h)	<i>E. coli</i>	
				(<i>E. coli</i> 100 mL ⁻¹)	(log ₁₀)
Background site	MS15	2007-03-28	9:15	< 10	1.0000
		2007-04-16	14:15	10	1.0000
		2007-06-26	12:30	100	2.0000
Downstream site for field F3	MS3	2007-03-22	14:10	580	2.7634
		2007-03-25	10:59	140	2.1461
		2007-03-26	9:40	50	1.6990
		2007-03-27	9:03	60	1.7782
		2007-03-28	10:35	20	1.3010
		2007-03-29	10:55	10	1.0000
		2007-04-16	9:55	30	1.4771
		2007-04-26	12:15	< 10	1.0000
		2007-05-22	15:10	15,000	4.1761
		2007-05-22	7:50	8,000	3.9031
		2007-05-23	10:00	11,200	4.0492
		2007-05-27	11:55	590	2.7709
		2007-05-28	13:00	70	1.8451
		2007-05-30	14:30	70	1.8451
		2007-05-31	13:10	67	1.8261
		2007-06-07	9:10	3,000	3.4771
		2007-06-07	16:15	820	2.9138
		2007-06-24	12:40	200	2.3010
		2007-06-25	11:10	600	2.7782
		2007-06-26	10:30	90	1.9542
		2007-07-10	9:30	1,500	3.1761

Field notes on livestock presence:

May 16 - 36 cows plus calves in riparian areas of fields F3 and F4 upstream of MS3 and MS4

May 26 - cattle moved from riparian areas F3 and F4

Aug. 4 - cattle moved into riparian areas F3 and F4 upstream of MS3 and MS4

Aug. 14 - cattle moved out of riparian areas F3 and F4

Appendix 4 continued. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) collected from the WEBs Stepler Watershed Project, Manitoba, Canada, 2007.

Sample site reference	Sample site	date (y/m/d)	time (h)	<i>E. coli</i>	
				(<i>E. coli</i> 100 mL ⁻¹)	(log ₁₀)
Downstream site for field F4	MS4	2007-03-25	10:50	110	2.0414
		2007-03-25	16:00	30	1.4771
		2007-03-26	9:30	90	1.9542
		2007-03-27	8:50	60	1.7782
		2007-03-28	10:25	10	1.0000
		2007-03-29	10:45	20	1.3010
		2007-04-16	9:25	< 10	1.0000
		2007-05-22	15:00	16,000	4.2041
		2007-05-22	7:40	6,600	3.8195
		2007-05-23	10:10	2,900	3.4624
		2007-05-27	12:00	800	2.9031
		2007-05-28	13:30	280	2.4472
		2007-05-31	13:00	250	2.3979
		2007-06-07	9:00	1,440	3.1584
		2007-06-07	16:00	300	2.4771
		2007-06-24	11:55	300	2.4771
		2007-06-25	11:00	400	2.6021
		2007-06-26	10:20	160	2.2041
		2007-07-10	9:10	610	2.7853

Field notes on livestock presence:

May 16 - 36 cows plus calves in riparian areas of fields F3 and F4 upstream of MS3 and MS4
 May 26 - cattle moved from riparian areas F3 and F4
 Aug. 4 - cattle moved into riparian areas F3 and F4 upstream of MS3 and MS4
 Aug. 14 - cattle moved out of riparian areas F3 and F4

Appendix 4 continued. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) collected from the WEBs Stepler Watershed Project, Manitoba, Canada, 2007.

Sample site reference	Sample site	date (y/m/d)	time (h)	<i>E. coli</i>	
				(<i>E. coli</i> 100 mL ⁻¹)	(log ₁₀)
Downstream site for farmyard- cattle-corral (holding pond inlet area)	MS5	2007-03-21	14:45	24,000	4.3802
		2007-03-25	11:50	overgrown	-
		2007-03-25	17:50	230,000	5.3617
		2007-03-26	9:15	4,300	3.6335
		2007-03-26	15:00	2,300	3.3617
		2007-03-27	9:20	21,000	4.3222
		2007-03-27	12:40	23,000	4.3617
		2007-03-28	10:45	38,000	4.5798
		2007-03-28	12:25	4,300	3.6335
		2007-03-29	9:10	43,000	4.6335
		2007-04-02	13:00	75,000	4.8751
		2007-04-16	10:32	430	2.6335
		2007-04-26	13:10	3	0.4771
		2007-05-06	12:45	46,000	4.6628
		2007-05-07	8:10	43	1.6335
		2007-05-07	14:36	< 3	0.4771
		2007-05-10	9:45	4	0.6021
		2007-05-22	12:55	430,000	5.6335
		2007-05-22	7:30	930,000	5.9685
		2007-05-23	8:35	2,300,000	6.3617
		2007-05-23	13:00	230,000	5.3617
		2007-05-27	10:35	43,000	4.6335
		2007-05-28	13:40	930	1.9685
		2007-05-30	12:45	93	1.9685
		2007-05-31	13:30	230	2.3617
		2007-06-06	18:12	43,000	4.6335
		2007-06-07	15:15	2,300,000	6.3617
		2007-06-07	9:55	930,000	5.9685
		2007-06-13	12:45	93,000	4.9685
		2007-06-18	8:05	93,000	4.9685
		2007-06-19	9:05	9,300	3.9685
		2007-06-20	13:15	380	2.5798
		2007-06-24	9:45	230,000	5.3617
		2007-06-25	8:00	930,000	5.9685
		2007-06-25	14:25	430,000	5.6335
		2007-06-26	8:15	930,000	5.9685
		2007-06-26	13:50	380,000	5.5798
		2007-06-27	12:40	210,000	5.3222
		2007-07-03	12:30	230	2.3617
		2007-07-05	13:00	23,000	4.3617
		2007-07-10	11:30	430,000	5.6335
		2007-07-12	13:20	9,300	3.9685

Appendix 4 continued. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) collected from the WEBs
 Stepler Watershed Project, Manitoba, Canada, 2007.

Sample site reference	Sample site	date (y/m/d)	time (h)	<i>E. coli</i>	
				(<i>E. coli</i> 100 mL ⁻¹)	(log ₁₀)
Holding pond near irrigation pump intake	MS5A	2007-04-02	13:00	7,500	3.8751
		2007-04-04	12:15	43,000	4.6335
		2007-04-09	12:30	28,000	4.4472
		2007-04-12	12:45	18,200	4.2601
		2007-04-16	10:40	15,000	4.1761
		2007-04-18	10:30	9,300	3.9685
		2007-04-18	17:00	4,300	3.6335
		2007-04-19	9:00	15,000	4.1761
		2007-04-19	14:00	15,000	4.1761
		2007-04-23	8:30	1,500	3.1761
		2007-04-23	15:00	430	2.6335
		2007-04-26	9:30	430	2.6335
		2007-04-26	13:15	150	2.1761
		2007-04-30	9:00	14	1.1461
		2007-04-30	15:00	15	1.1761
		2007-05-02	8:55	7	0.8451
		2007-05-03	8:30	9	0.9542
		2007-05-03	14:50	< 3	0.4771
		2007-05-07	8:20	43	1.6335
		2007-05-07	8:20	4	0.6021
		2007-05-10	9:50	< 3	0.4771
		2007-05-10	13:05	< 3	0.4771
		2007-05-27	10:35	93,000	4.9685
		2007-05-30	12:45	23	1.3617
		2007-06-04	8:20	200	2.3010
		2007-06-04	15:35	200	2.3010
		2007-06-05	8:30	290	2.4624
		2007-06-12	12:30	93,000	4.9685
		2007-06-20	9:55	2,100	3.3222
		2007-06-21	5:00	4,300	3.6335
		2007-06-21	13:00	9,300	3.9685
		2007-07-03	12:00	750	2.8751
		2007-07-09	12:05	2,300	3.3617
		2007-07-17	12:15	4,300	3.6335
		2007-07-19	13:44	930	2.9685
		2007-08-01	11:00	2,300	3.3617
		2007-08-02	11:00	4,300	3.6335

Appendix 4 continued. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) collected from the WEBs Steppler Watershed Project, Manitoba, Canada, 2007.

Sample site reference	Sample site	date (y/m/d)	time (h)	<i>E. coli</i>	
				(<i>E. coli</i> 100 mL ⁻¹)	(log ₁₀)
Holding pond spillway	MS5B	2007-06-25	10:35	9,300	3.9685
		2007-06-25	14:30	9,300	3.9685
		2007-06-25	8:05	4,300	3.6335
		2007-06-26	8:20	430,000	5.6335
		2007-06-26	13:50	230,000	5.3617
		2007-06-27	9:05	43,000	4.6335
		2007-06-27	12:40	230,000	5.3617
		2007-06-28	12:35	43,000	4.6335
Irrigation sprinkler nozzle discharge	MS5C	2007-04-23	15:00	2,300	3.3617
		2007-04-26	9:30	150	2.1761
		2007-04-26	13:15	150	2.1761
		2007-04-30	15:00	38	1.5798
		2007-05-03	14:50	4	0.6021
		2007-05-07	14:30	9	0.9542
		2007-07-03	12:15	1,500	3.1761

Appendix 4 continued. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) collected from the WEBs Stepler Watershed Project, Manitoba, Canada, 2007.

Sample site reference	Sample site	date (y/m/d)	time (h)	<i>E. coli</i>	
				(<i>E. coli</i> 100 mL ⁻¹)	(log ₁₀)
Downstream site of MS3, MS4, and MS5	MS7	2007-03-25	12:15	93	1.9685
		2007-03-25	16:55	120	2.0792
		2007-03-26	11:40	150	2.1761
		2007-03-26	15:15	23	1.3617
		2007-03-27	9:57	9	0.9542
		2007-03-27	13:40	43	1.6335
		2007-03-28	10:55	< 10	1.0000
		2007-03-29	9:25	50	1.6990
		2007-04-16	13:00	< 1	0.0000
		2007-04-26	12:30	30	1.4771
		2007-05-07	13:15	60	1.7782
		2007-05-22	13:35	32,000	4.5051
		2007-05-22	8:15	54,000	4.7324
		2007-05-23	9:10	1,140	3.0569
		2007-05-27	11:10	210	2.3222
		2007-05-28	8:20	280	2.4472
		2007-05-28	14:00	190	2.2788
		2007-05-30	13:30	130	2.1139
		2007-05-31	13:45	330	2.5185
		2007-06-05	9:40	20	1.3010
		2007-06-06	18:25	2,900	3.4624
		2007-06-07	9:40	9,000	3.9542
		2007-06-07	14:45	11,900	4.0755
		2007-06-18	8:55	210	2.3222
		2007-06-24	10:20	200	2.3010
		2007-06-25	10:00	900	2.9542
		2007-06-25	14:05	2,000	3.3010
		2007-06-26	9:00	700	2.8451
		2007-06-27	13:05	10	1.0000
		2007-06-28	14:00	10	1.0000
		2007-07-03	12:40	200	2.3010
		2007-07-10	11:05	2,600	3.4150

Field notes on livestock presence:

May 16 - cattle west side of Field F5 upstream of MS7
 May 22 - cattle on west side of water course in Field F5 upstream of MS7
 May 26 - cattle moved from west side to east side of Field F5
 May 30 - cattle in east side of Field F5 and immediately upstream of MS7
 June 1 - cattle moved out of WEBs area
 Aug. 4 - cattle moved into west side of Field F5 that is ~ 500 m upstream of MS7
 Aug. 14 - cattle moved to east side of Field F5 that is near MS7
 Aug. 21 - cattle moved out of Field F5

Appendix 4 continued. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) collected from the WEBs
 Stepler Watershed Project, Manitoba, Canada, 2007.

Sample site reference	Sample site	date (y/m/d)	time (h)	<i>E. coli</i>	
				(<i>E. coli</i> 100 mL ⁻¹)	(log ₁₀)
Downstream site from MS7 and MS6	MS9	2007-03-25	12:30	30	1.4771
		2007-03-25	17:00	110	2.0414
		2007-03-26	15:25	23	1.3617
		2007-03-26	12:00	23	1.3617
		2007-03-27	10:10	9	0.9542
		2007-03-27	13:15	4	0.6021
		2007-03-28	11:10	< 10	1.0000
		2007-03-29	9:55	30	1.4771
		2007-04-16	13:15	< 1	0.0000
		2007-04-26	12:45	< 1	0.0000
		2007-05-07	13:30	170	2.2304
		2007-05-22	13:55	16,000	4.2041
		2007-05-22	8:25	290,000	5.4624
		2007-05-23	9:30	1,700	3.2304
		2007-05-27	11:30	20	1.3010
		2007-05-28	14:30	240	2.3802
		2007-05-30	13:45	190	2.2788
		2007-05-31	14:00	100	2.0000
		2007-06-05	9:30	70	1.8451
		2007-06-07	9:50	880	2.9445
		2007-06-07	14:00	4,000	3.6021
		2007-06-18	9:10	330	2.5185
		2007-06-24	10:45	400	2.6021
		2007-06-25	10:10	900	2.9542
		2007-06-25	13:35	500	2.6990
		2007-06-26	9:15	700	2.8451
		2007-06-27	13:20	20	1.3010
		2007-06-28	14:05	40	1.6021
		2007-07-03	12:45	60	1.7782
		2007-07-10	10:10	2,500	3.3979

Appendix 4 continued. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) collected from the WEBs
 Stepler Watershed Project, Manitoba, Canada, 2007.

Sample site reference	Sample site	date (y/m/d)	time (h)	<i>E. coli</i>	
				(<i>E. coli</i> 100 mL ⁻¹)	(log ₁₀)
Steppler Dam outlet	MS12	2007-03-25	12:45	20	1.3010
		2007-03-25	17:05	50	1.6990
		2007-03-26	12:00	90	1.9542
		2007-03-26	15:30	50	1.6990
		2007-03-27	10:20	20	1.3010
		2007-03-27	13:17	10	1.0000
		2007-03-28	11:20	10	1.0000
		2007-03-29	10:05	< 10	1.0000
		2007-04-16	13:15	10	1.0000
		2007-05-22	14:15	10	1.0000
		2007-05-22	8:25	100	2.0000
		2007-05-23	9:30	1,480	3.1703
		2007-05-27	11:35	20	1.3010
		2007-05-30	13:45	< 1	0.0000
		2007-05-31	13:55	6	0.7782
		2007-06-07	9:45	< 1	0.0000
		2007-06-07	13:50	80	1.9031
		2007-06-18	9:05	120	2.0792
		2007-06-24	10:55	160	2.2041
		2007-06-25	9:10	350	2.5441
		2007-06-26	9:10	110	2.0414
		2007-06-27	13:15	40	1.6021
		2007-07-10	9:45	190	2.2788

Appendix 4 continued. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) collected from the WEBs Stepler Watershed Project, Manitoba, Canada, 2007.

Sample site reference	Sample site	date (y/m/d)	time (h)	<i>E. coli</i>	
				(<i>E. coli</i> 100 mL ⁻¹)	(log ₁₀)
Upstream ditch site from winter holding area	MS16	2007-03-13	8:40	830	2.9191
		2007-03-13	15:00	550	2.7404
		2007-03-14	12:10	190	2.2788
		2007-03-19	8:15	80	1.9031
		2007-03-21	12:30	80	1.9031
		2007-03-21	16:40	100	2.0000
		2007-03-22	13:35	< 10	1.0000
		2007-03-25	11:25	15	1.1761
		2007-05-22	9:40	500	2.6990
		2007-05-23	8:40	170	2.2304
		2007-05-27	13:30	50	1.6990
		2007-06-06	18:00	387,000	5.5877
		2007-06-07	15:40	177,000	5.2480
		2007-06-13	12:45	12,300	4.0899
		2007-06-18	8:40	20,400	4.3096
		2007-06-24	10:00	13,000	4.1139
		2007-06-25	8:55	63,000	4.7993
		2007-06-26	11:10	8,700	3.9395
		2007-07-10	9:45	5,600	3.7482
Downstream ditch site from winter holding area	MS17	2007-03-13	8:15/15:	both overgrown	-
		2007-03-14	12:20	30	1.4771
		2007-03-19	8:20	4	0.6021
		2007-03-19	8:20	7	0.8451
		2007-03-21	12:45	43	1.6335
		2007-03-21	16:45	43	1.6335
		2007-03-22	13:25	9	0.9542
		2007-03-25	12:05	34	1.5315
		2007-05-22	8:10	430,000	5.6335
		2007-05-23	8:40	30	1.4771
		2007-05-27	13:30	< 1	0.0000
		2007-06-06	18:00	32,000	4.5051
		2007-06-07	15:40	15,400	4.1875
		2007-06-13	12:45	500	2.6990
		2007-06-18	8:45	1,400	3.1461
		2007-06-24	10:50	900	2.9542
		2007-06-25	9:00	6,000	3.7782
		2007-06-26	11:20	540	2.7324
		2007-07-10	9:50	1,200	3.0792

Field notes on livestock presence:

June 6 - sheep in pasture adjacent and u/s to MS16

June 13 - sheep in pasture on west side of lane further from but still upstream of MS16

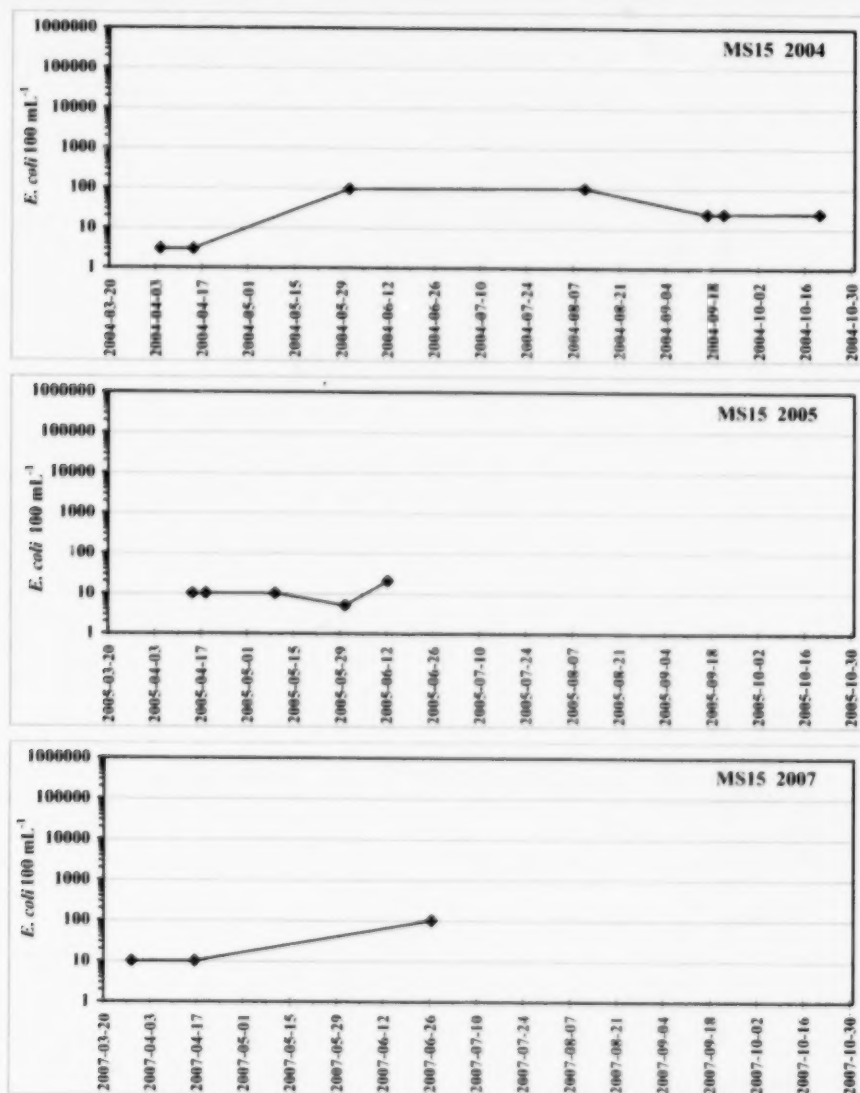
June 18 - sheep moved to east side of lane immediately upstream of MS16

June 24 - sheep moved to west side of lane further away but upstream of MS16

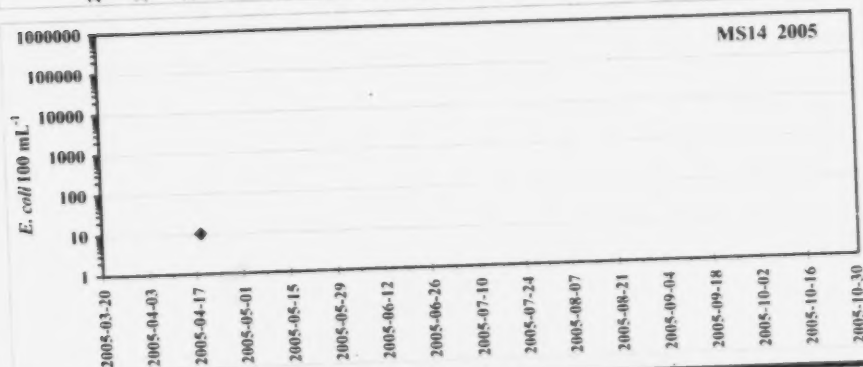
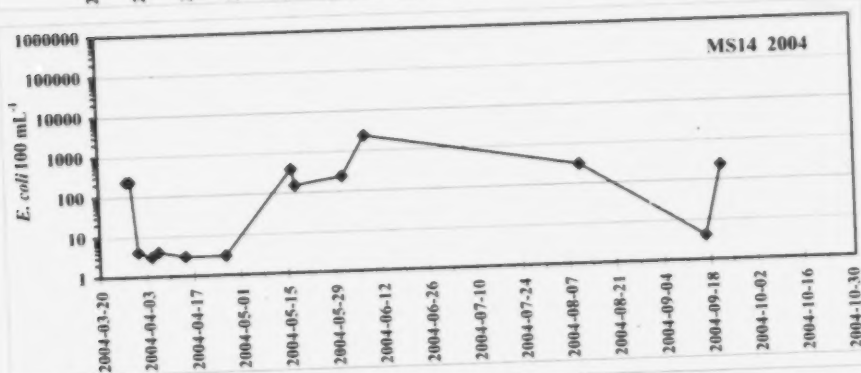
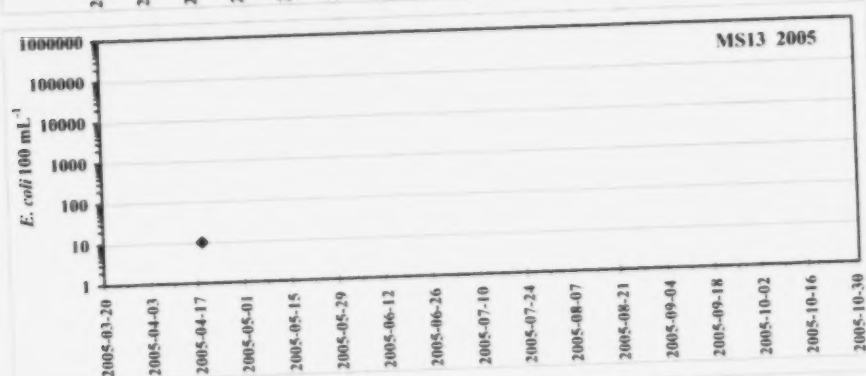
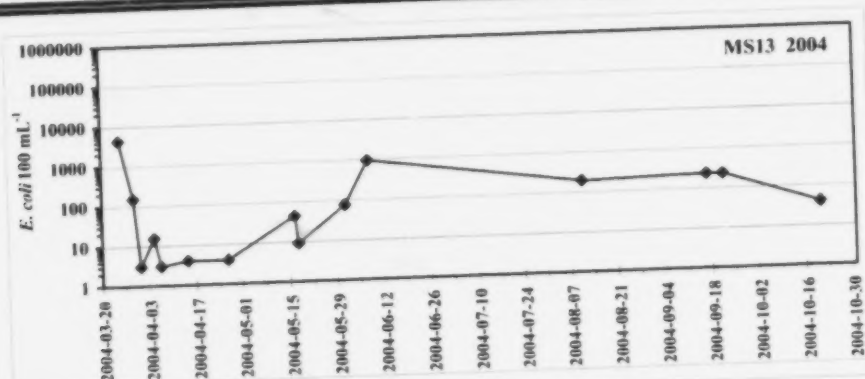
Appendix 4 continued. *Escherichia coli* counts (*E. coli* 100 mL⁻¹) collected from the WEBs Steppler Watershed Project, Manitoba, Canada, 2007.

Sample site reference	Sample site	date (y/m/d)	time (h)	<i>E. coli</i>	
				(<i>E. coli</i> 100 mL ⁻¹)	(log ₁₀)
Confined area drainage ditch test-well #1	DTW1	2007-05-30	12:55	< 1	0.0000
		2007-06-19	9:55	50	1.6990
		2007-06-26	9:00	10	1.0000
Confined area drainage ditch test-well #2	DTW2	2007-05-03	14:15	300	2.4771
		2007-05-07	14:08	10	1.0000
		2007-05-22	13:10	16,000	4.2041
		2007-05-30	13:00	50	1.6990
		2007-06-19	10:05	30	1.4771
		2007-06-26	9:15	20	1.3010
		2007-08-01	14:05	100	2.0000
Confined area drainage ditch test-well #3	DTW3	2007-04-04	11:30	15	1.1761
		2007-04-19	14:15	10	1.0000
		2007-04-26	13:00	< 1	0.0000
		2007-05-07	13:50	< 1	0.0000
		2007-05-30	13:10	10	1.0000
		2007-06-19	9:30	1,450	3.1614
		2007-06-26	8:45	270	2.4314
		2007-08-01	14:30	200	2.3010
Confined area drainage ditch test-well #4	DTW4	2007-04-04	11:45	150,000	5.1761
		2007-04-19	14:30	10,500	4.0212
		2007-04-26	13:05	2,100	3.3222
		2007-05-03	14:00	1,130	3.0531
		2007-05-07	14:00	880	2.9445
		2007-05-22	13:10	33,000	4.5185
		2007-05-30	13:15	57,000	4.7559
		2007-06-19	9:40	11,100	4.0453
		2007-06-26	8:50	34,000	4.5315
Borehole test-well #9	BH9	2007-05-03	14:30	< 1	0.0000
Borehole test-well #11	BH11	2007-05-03	14:30	< 1	0.0000

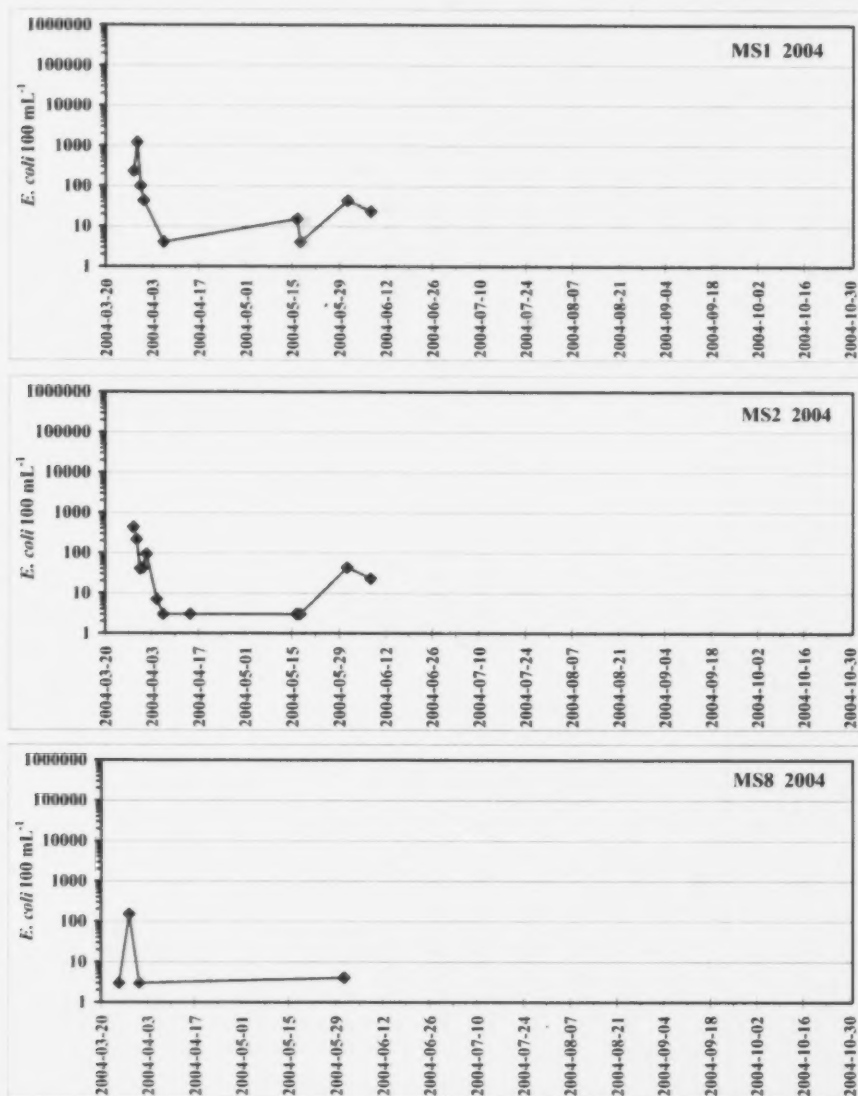
Appendix 5a. Graphs showing daily *E. coli* counts (logarithmic scale) for site MS15 of the WEBS Stepper Watershed Project, Manitoba, Canada, 2004 to 2007.



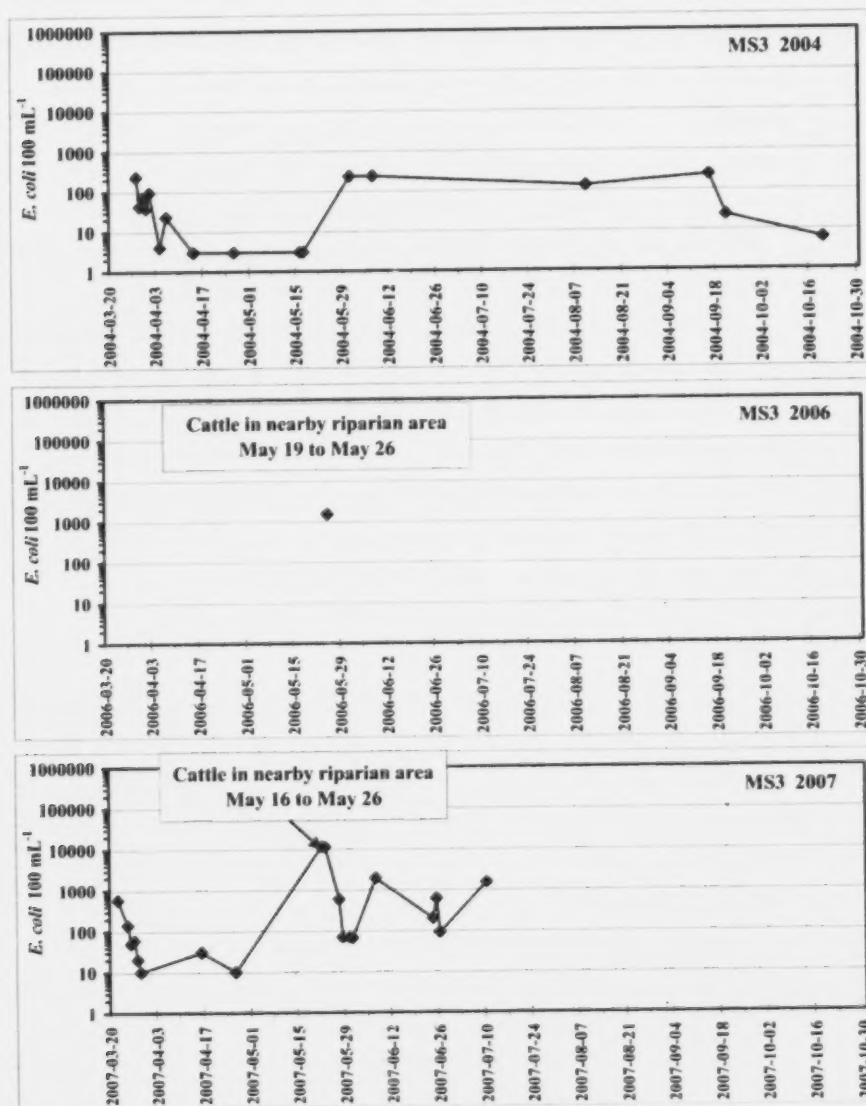
Appendix 5b. Graphs showing daily *E. coli* counts (log scale) for sites MS13 and MS14 of the WEBS Stepper Watershed Project, Manitoba, Canada, 2004 to 2007.



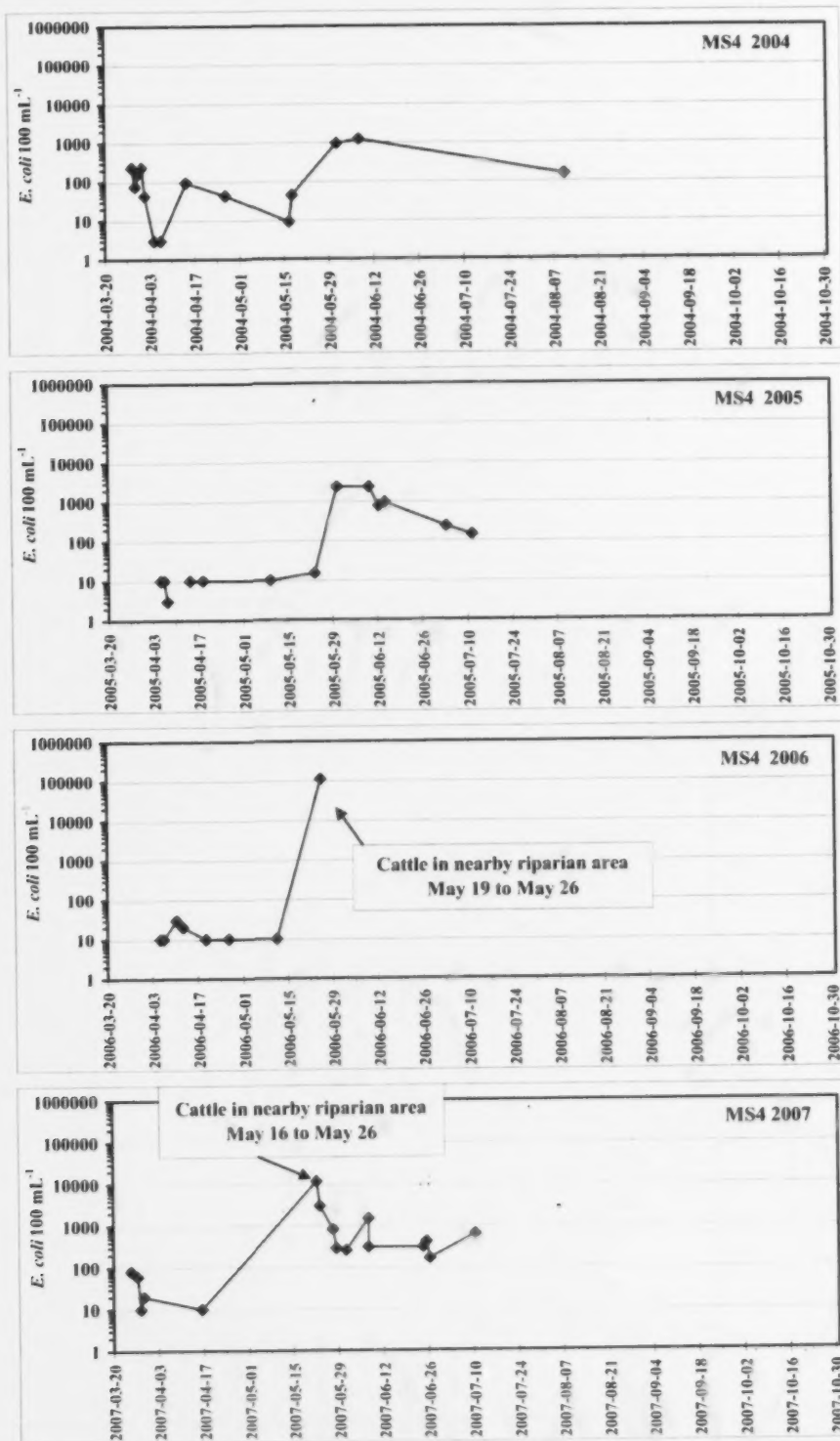
Appendix 5c. Graphs showing daily *E. coli* counts (logarithmic scale) for sites MS1, MS2, and MS8 of the WEBs Stepper Watershed Project, Manitoba, Canada, 2004 to 2007.



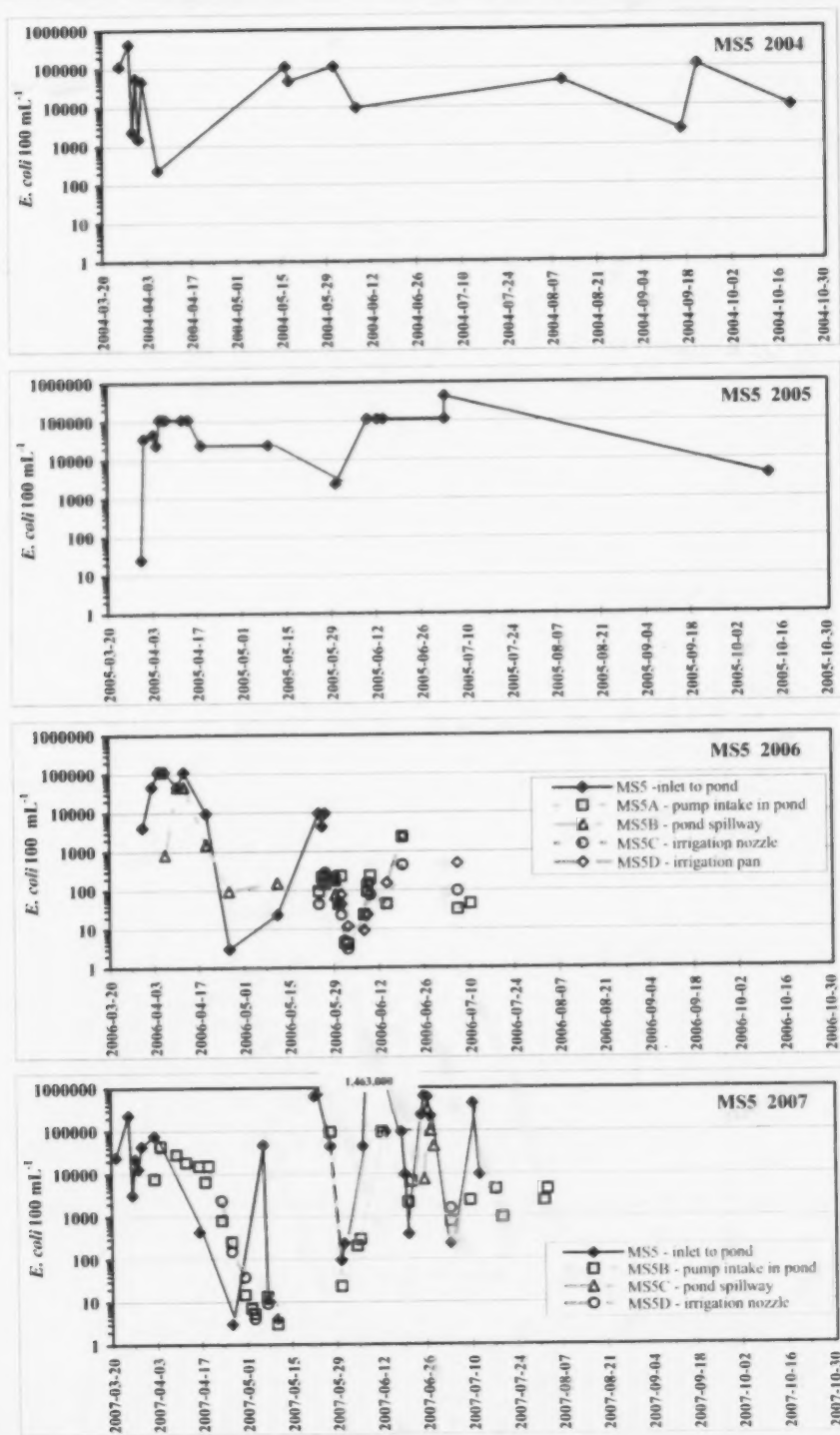
Appendix 5d. Graphs showing daily *E. coli* counts (logarithmic scale) for site MS3 of the WEBs Stepper Watershed Project, Manitoba, Canada, 2004 to 2007.



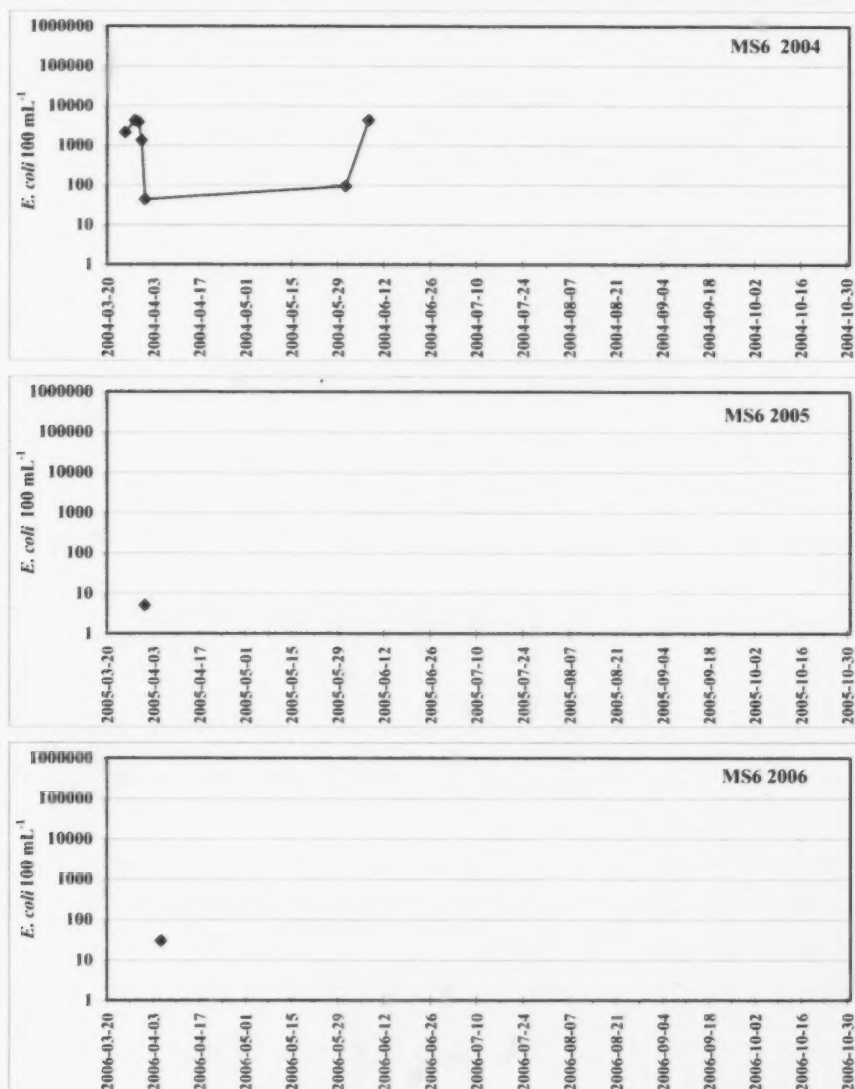
Appendix 5c. Graphs showing daily *E. coli* counts (logarithmic scale) for site MS4 of the WEBs Stepper Watershed Project, Manitoba, Canada, 2004 to 2007.



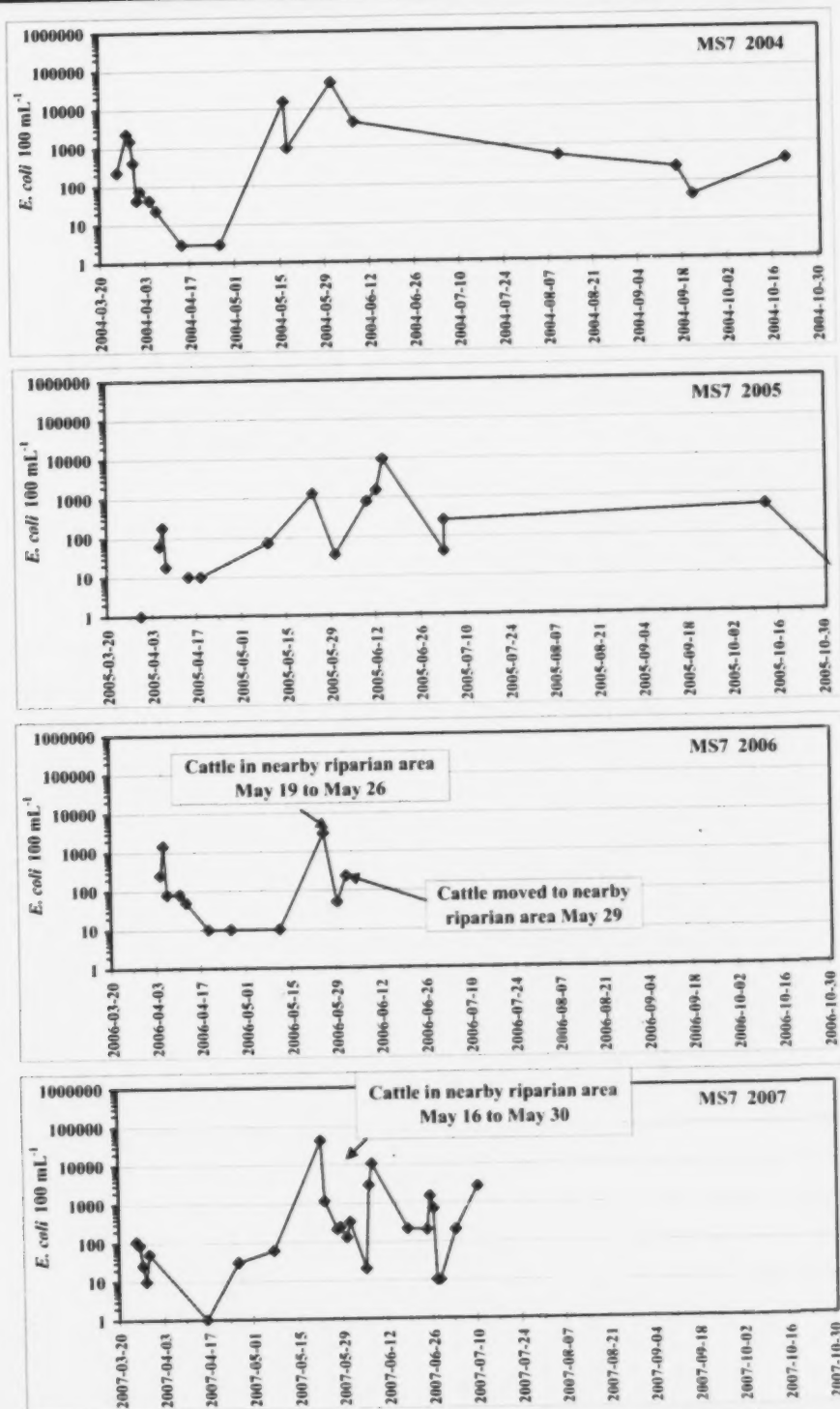
Appendix 5f. Graphs showing daily *E. coli* counts (logarithmic scale) for site MS5 of the WEBs Stepper Watershed Project, Manitoba, Canada, 2004 to 2007.



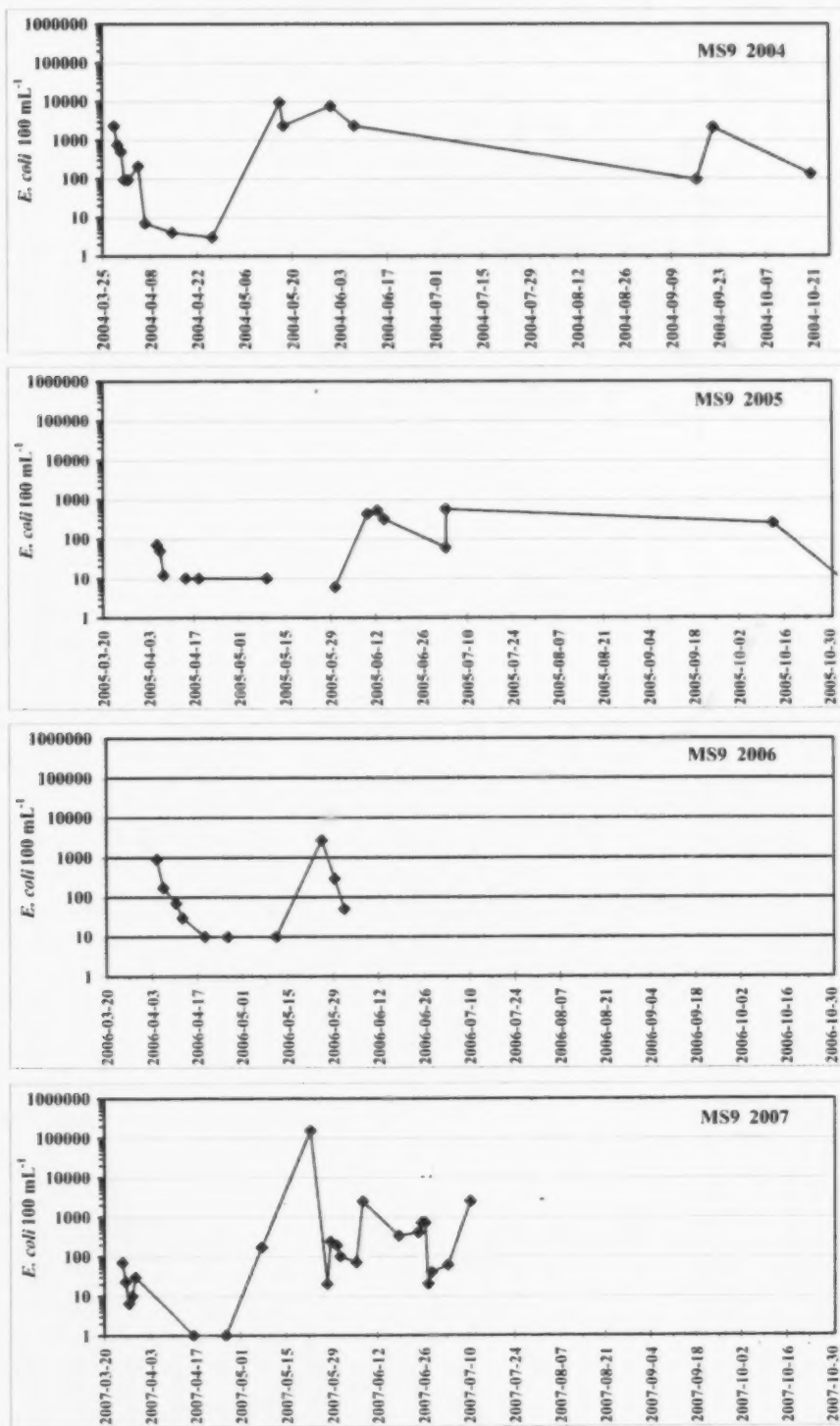
Appendix 5g. Graphs showing daily *E. coli* counts (logarithmic scale) for site MS6 of the WEBS Stepper Watershed Project, Manitoba, Canada, 2004 to 2007.



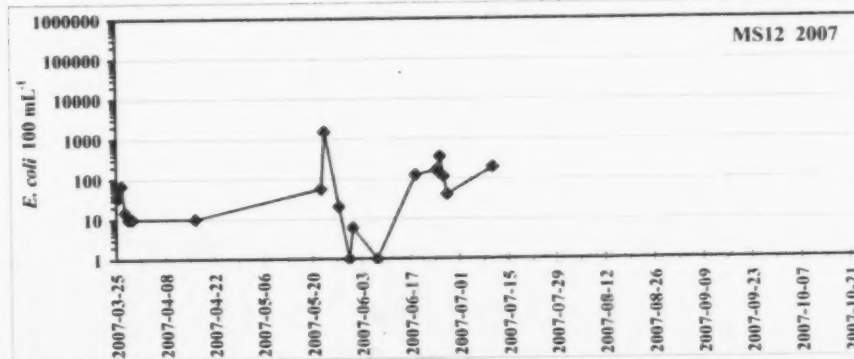
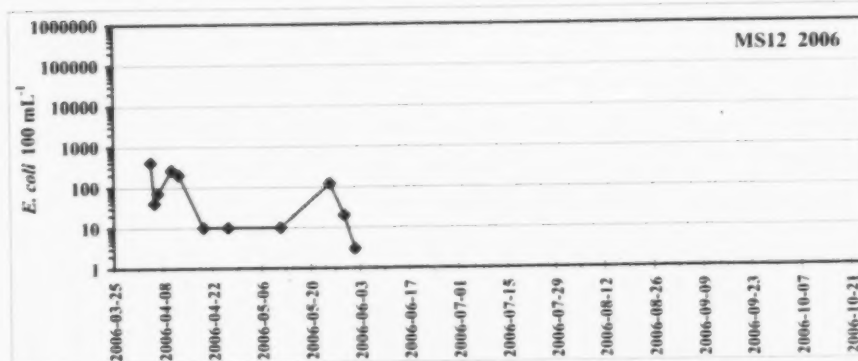
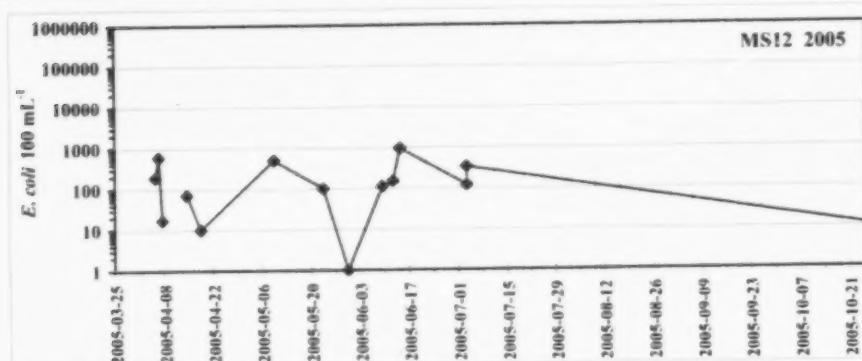
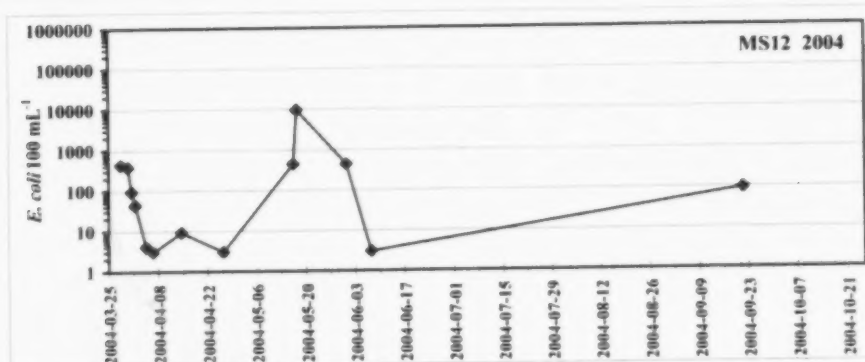
Appendix 5h. Graphs showing daily *E. coli* counts (logarithmic scale) for site MS7 of the WEBs Stepper Watershed Project, Manitoba, Canada, 2004 to 2007.



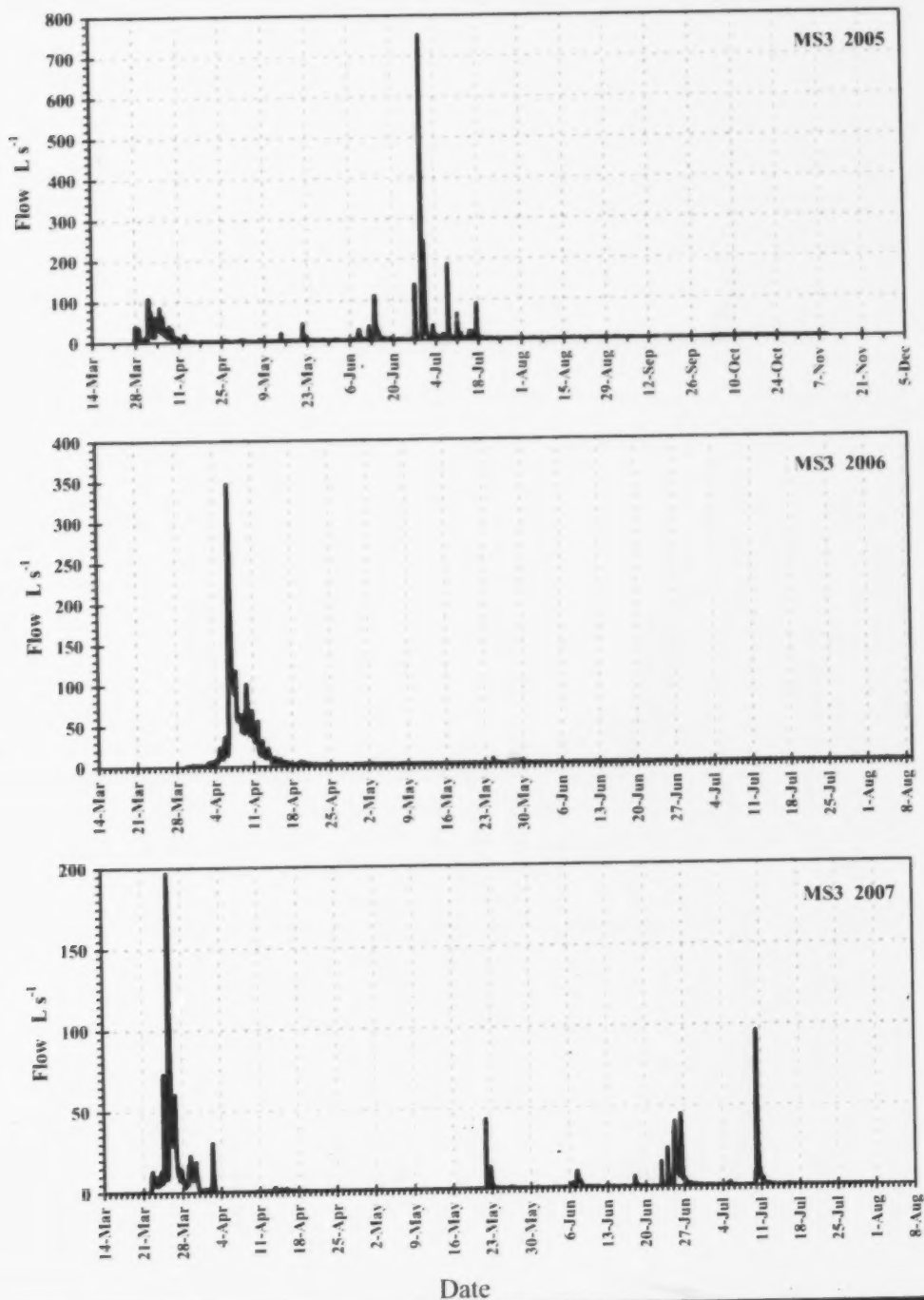
Appendix 5i. Graphs showing daily *E. coli* counts (logarithmic scale) for site MS9 of the WEBS Stepper Watershed Project, Manitoba, Canada, 2004 to 2007.



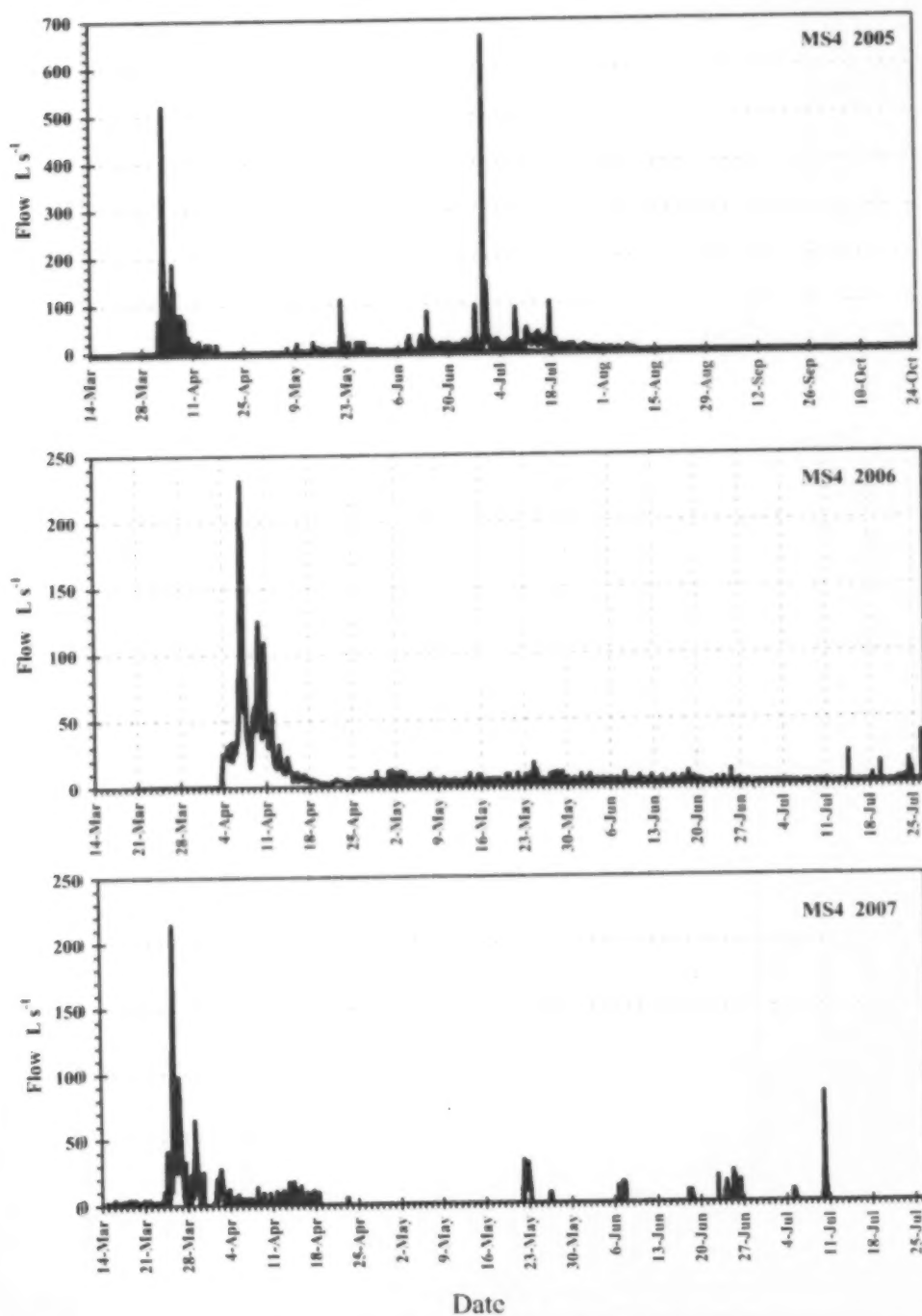
Appendix 5j. Graphs showing daily *E. coli* counts (logarithmic scale) for site MS12 of the WEBs Stepper Watershed Project, Manitoba, Canada, 2004 to 2007.



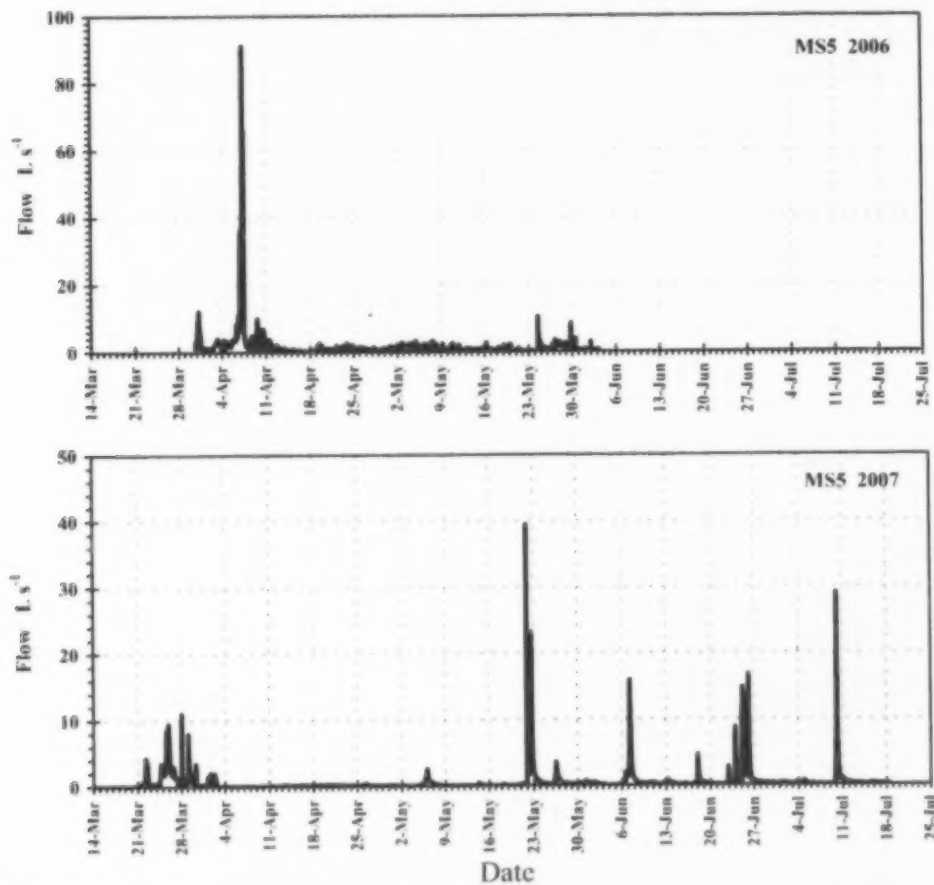
Appendix 6a. Annual hydrographs for site MS3 in the WEBs Steppler Watershed Project, Manitoba, Canada, 2004 to 2007. Note: Hydrology information supplied by J. Yarotski, PFRA, Regina.



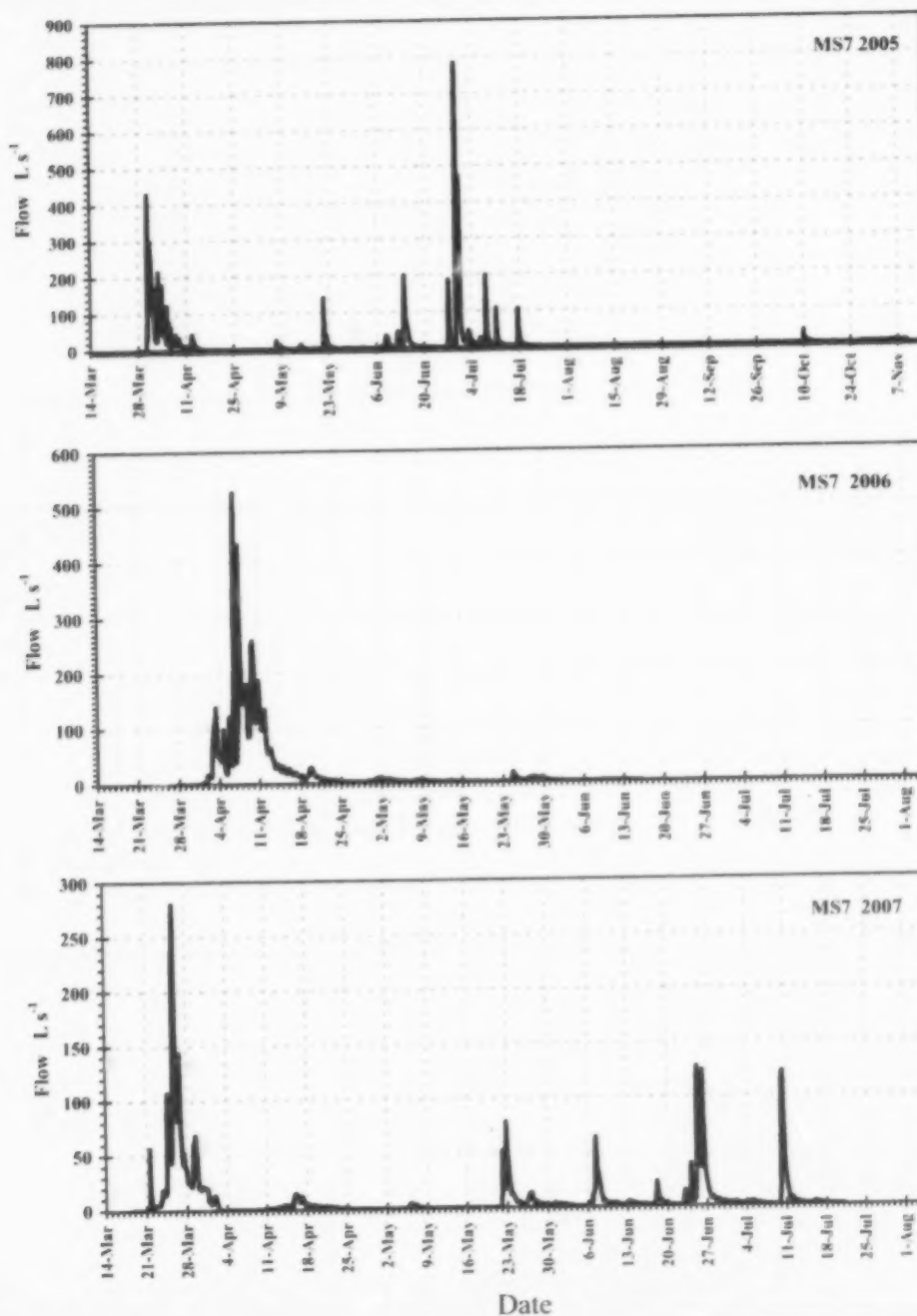
Appendix 6b. Annual hydrographs for site MS4 in the WEBs Stepples Watershed Project, Manitoba, Canada, 2004 to 2007. Note: Hydrology information supplied by J. Yarotski, PFRA, Regina.



Appendix 6c. Annual hydrographs for site MS5 in the WEBs Stepipler Watershed Project, Manitoba, Canada, 2004 to 2007. Note: Hydrology information supplied by J. Yarotski, PFRA, Regina.



Appendix 6d. Annual hydrographs for site MS7 in the WEBs Steppler Watershed Project, Manitoba, Canada, 2004 to 2007. Note: Hydrology information supplied by J. Yarotski, PFRA, Regina.



Appendix 7. Pairwise comparisons ($\alpha = 0.05$) of mean *E. coli* counts between sampling sites in the WEBs Steppeler Watershed Project, Manitoba, Canada, 2004 to 2007.
Note: p-values of each comparison is shown within each box.

Year		Steppeler Watershed sites										Background sites		
		MS1	MS3	MS2	MS4	MS5	MS7	MS6	MS9	MS8	MS12	MS13	MS14	MS15
2004	MS1	1.000												
	MS3	1.000	1.000											
	MS2	1.000	1.000	1.000										
	MS4	1.000	0.999	0.999	1.000									
	MS5	0.000	0.000	0.000	0.000	1.000								
	MS7	0.926	0.312	0.378	0.923	0.002	1.000							
	MS6	0.551	0.109	0.134	0.529	0.394	0.998	1.000						
	MS9	0.938	0.354	0.421	0.938	0.003	1.000	0.998	1.000					
	MS8	0.996	1.000	1.000	0.982	0.000	0.406	0.152	0.431	1.000				
	MS12	1.000	0.981	0.988	1.000	0.000	0.992	0.761	0.994	0.933	1.000			
	MS13	1.000	1.000	1.000	1.000	0.000	0.483	0.180	0.525	1.000	0.995	1.000		
	MS14	1.000	1.000	1.000	1.000	0.000	0.627	0.256	0.666	1.000	0.999	1.000	1.000	
	MS15	0.988	1.000	1.000	0.960	0.000	0.316	0.111	0.339	1.000	0.884	1.000	0.999	1.000
2005							MS4	MS5	MS7	MS9	MS12	MS15		
	MS4						1.000							
	MS5						0.000	1.000						
	MS7						0.984	0.000	1.000					
	MS9						1.000	0.000	0.971	1.000				
	MS12						0.870	0.000	0.997	1.000	1.000			
	MS15						0.988	0.000	0.840	0.995	0.638	1.000		
2006							MS4	MS5	MS7	MS9	MS12			
	MS4						1.000							
	MS5						0.001	1.000						
	MS7						0.976	0.004	1.000					
	MS9						0.975	0.006	1.000	1.000				
	MS12						0.999	0.001	0.994	0.992	1.000			
2007							MS3	MS4	MS5	MS7	MS9	MS12		
	MS3						1.000							
	MS4						1.000	1.000						
	MS5						0.027	0.055	1.000					
	MS7						1.000	0.999	0.005	1.000				
	MS9						0.959	0.941	0.001	0.991	1.000			
	MS12						0.429	0.406	0.000	0.542	0.877	1.000		

 = significant difference at $p < 0.05$

 = no significant difference

Appendix 8. Least squares regression analyses on *E. coli* counts within the Stepler Watershed holding pond after the initial spring runoff period ended, 2006 and 2007.

Year	n	equation	r ²	p
2006	3	Log ₁₀ <i>E. coli</i> = 4.7452 – 2.1295 (log ₁₀ days)	0.9434	0.153
2007	17	Log ₁₀ <i>E. coli</i> = 7.3701 – 3.6617 (log ₁₀ days)	0.555	0.0006

Appendix 9. Secchi disk measurements collected during 2007 from the holding pond capturing runoff from the farmyard-cattle-corral in the WEBs Stepler Watershed Project, Manitoba, Canada, 2004 to 2007.

Date	Secchi disk measurement (cm)
May 31	12 – 13
June 4	19
June 5	15
June 7	16
June 8	13.5
June 12	7
June 13	10
June 18	22
June 20	22
June 29	17
July 5	17
July 30	14
August 2	14

